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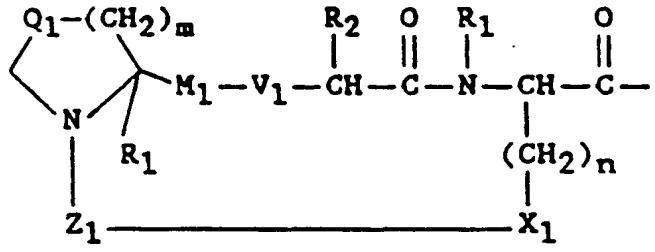

(57) Abstract

The present invention provides novel peptides. These peptides are useful as renin inhibitors and have a novel cyclic tripeptide of formula (I) at the N-terminus. Some of these peptides also have a non-cleavable transition state insert corresponding to the 10,11-position of the renin substrate (angiotensinogen). Renin inhibitors are useful for the diagnosis and control of renin-dependent hypertension, congestive heart failure, renin-dependent hyperaldosterism, other renin-dependent cardiovascular disorders and ocular disorders. These peptides are also useful as inhibitors of HIV-I protease. Inhibitors of HIV-I protease are useful for treating humain acquired immunodeficiency disease syndrome (AIDS).

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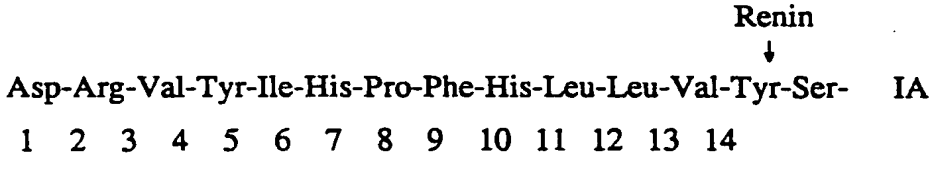
PEPTIDES CONTAINING CYCLIC TRIPEPTIDES

DESCRIPTION

BACKGROUND OF THE INVENTION

The present invention provides novel compounds. More particularly, the present invention provides novel peptide analogs. The peptides of the present invention are useful as renin-inhibitory peptides and contain a novel cyclic tripeptide at the N-terminus of the peptide. Renin inhibitors are useful for the diagnosis and control of renin-dependent hypertension, congestive heart failure, renin-dependent hyperaldosterism, and other renin-dependent cardiovascular disorders. The peptides of the present invention are also useful as inhibitors of retroviral proteases. Inhibitors of retroviral proteases, such as the HIV-I protease, are useful for treating diseases caused by retroviruses, such as human acquired immunodeficiency disease syndrome (AIDS).

Renin is an endopeptidase which specifically cleaves a particular peptide bond of its substrate (angiotensinogen), of which the N-terminal sequence in equine substrate is for example:



IA

as found by L.T. Skeggs, et al., J. Exper. Med. 106, 439 (1957). Human renin substrate has a different sequence as recently discovered by D.A. Tewkesbury, et al., Biochem. Biophys. Res. Comm. 99, 1311 (1981). It may be represented as follows:

IB

and having the sequence to the left of the arrow (↓) being as designated in formula IA above.

Renin cleaves angiotensinogen to produce angiotensin I, which is converted to the potent pressor angiotensin II. A number of angiotensin I converting enzyme inhibitors are known to be useful in the treatment of hypertension. Inhibitors of renin are also

useful in the treatment of hypertension.

A number of renin-inhibitory peptides have been disclosed. Thus, U.S. patent 4,424,207; European published applications 45,665; 104,041; and 156,322; and U.S. patent application, Serial No. 825,250, filed 3 February 1986; disclose certain peptides with the dipeptide at the 10,11-position containing an isostere bond. A number of statine derivatives stated to be renin inhibitors have been disclosed, see, e.g., European published applications 77,028; 81,783; 114,993; 156,319; and 156,321; and U.S. patents 4,478,826; 4,470,971; 4,479,941; and 4,485,099. Terminal disulfide cycles have also been disclosed in renin inhibiting peptides; see, e.g., U.S. patents 4,477,440 and 4,477,441. Aromatic and aliphatic amino acid residues at the 10,11 position of the renin substrate are disclosed in U.S. patents 4,478,827 and 4,455,303. C-terminal amide cycles are disclosed in U.S. patent 4,485,099 and European published applications 156,320 and 156,318. Certain tetrapeptides are disclosed in European publications 111,266 and 77,027. Further, European published application No. 118,223 discloses certain renin inhibiting peptide analogs where the 10-11 peptide link is replaced by a one to four atom carbon or carbon-nitrogen link. Additionally, Holladay, et al., in "Synthesis of Hydroxyethylene and Ketomethylene Dipeptide Isosteres", Tetrahedron Letters, Vol. 24, No. 41, pp. 4401-4404, 1983 disclose various intermediates in a process to prepare stereo-directed "ketomethylene" and "hydroxyethylene" dipeptide isosteric functional groups disclosed in the above noted U.S. Patent No. 4,424,207.

Additionally, published European Applications 45,161 and 53,017 disclose amide derivatives useful as inhibitors of angiotensin converting enzymes.

Certain dipeptide and tripeptides are disclosed in U.S. patents 4,514,332; 4,510,085; and 4,548,926 as well as in European published applications 128,762; 152,255; and 181,110. Pepstatin derived renin inhibitors have been disclosed in U.S. patent 4,481,192. Retro-inverso bond modifications at positions 10-11 have been disclosed in U.S. patent 4,560,505 and in European published applications 127,234 and 127,235. Derivatives of isosteric bond replacements at positions 10-11 have been disclosed in European published applications 143,746 and 144,209; and U.S. patent application, Serial No. 833,993, filed 27 February 1986. Isosteric bond modifications at positions 11-12 and 12-13 have been disclosed in European published application 179,352. Certain peptides containing 2-substituted statine analogues have been disclosed in European published application 157,409. Certain peptides containing 3-aminodeoxy-

statine have been disclosed in European published application 161,588. Certain peptides containing 1-amino-2-hydroxybutane derivatives at positions 10-11 have been disclosed in European published application 172,346. Certain peptides containing 1-amino-2-hydroxypropane derivatives at positions 10-11 have been disclosed in European published application 172,347. Certain peptides containing N-terminal amide cycles have been disclosed in U.S. patent application, Serial No. 844,716, filed 27 March 1986. Certain peptides containing dihalostatine have been disclosed in PCT application, Serial No. 000,713, filed 7 April 1986.

European published applications 156,322; 114,993; and 118,223; and U.S. patent application, Serial No. 798,459, filed 15 November 1985; U.S. patent application, Serial No. 825,250, filed 3 February 1986; U.S. patent application, Serial No. 833,993, filed 27 February 1986; and U.S. patent application, Serial No. 844,716, filed 27 March 1986; disclose hydroxamic acids or esters at the C-terminus.

INFORMATION DISCLOSURE

A number of cyclic tetrapeptides are disclosed in Chemical Abstracts. These peptides may be divided into three generic formulas: formula XXX, formula XL and formula L. The definitions of the variables in these formulas are apparent from the Chemical Abstracts structures. The following are examples of peptides which are included within the generic formula XXX and which are identified by their Chemical Abstracts Registry Numbers: 118984-46-6; 114195-50-5; 114195-49-2; 114195-47-0; 110114-88-0; 109121-93-9; 109121-92-8; 109121-91-7; 109121-90-6; 109075-09-4; 108146-59-4; 108072-00-0; 108071-95-0; 107982-61-6; 107982-60-5; 106973-32-4; 106894-13-7; 104994-59-4; 104849-14-1; 104849-13-0; 104849-12-9; 104849-07-2; 104849-06-1; 104849-05-0; 104849-04-9; 104848-98-8; 104848-97-7; 104848-96-6; 104848-95-5; 103487-50-9; 98568-94-6; 97644-10-5; 96894-68-7; 96090-02-7; 92050-62-9; 90965-62-1; 89890-87-9; 89826-01-7; 89760-53-2; 89760-52-1; 89363-03-1; 89363-01-9; 89311-47-7; 89311-46-6; 89311-45-5; 89311-44-4; 89311-43-3; 89311-42-2; 89311-41-1; 89278-33-1; 88927-75-7; 88927-74-6; 88643-73-6; 88643-28-1; 88568-94-9; 87219-41-8; 85838-52-4; 85800-05-1; 84366-82-5; 84255-86-7; 84107-30-2; 83829-20-3; 83751-10-4; 83797-39-1; 83209-65-8; 82081-23-0; 82067-64-9; 81391-65-3; 81017-85-8; 77100-18-6; 77087-68-4; 77087-67-3; 76710-65-1; 73759-07-6; 72029-16-4; 72029-15-3; 71996-70-8; 71996-69-5; 71996-65-1; 71996-64-0; 70812-31-6; 70779-88-3; 70779-87-2; 68889-97-4; 68857-04-5; 66447-52-7; 64295-

20-1; 64070-02-6; 64044-83-3; 56144-84-4; 55332-74-6; 53532-93-7; 53342-16-8; 52574-64-8; 52574-63-7; 52574-62-6; and 52574-61-5.

The following are examples of peptides which are included within the generic formula XL and which are identified by their Chemical Abstracts Registry Numbers: 58855-91-7; 19293-32-4; 15005-62-6; 14168-13-9; 14074-75-0; 14074-74-9; 14074-73-8; 14074-72-7; 14051-89-9; 14051-88-8; 15920-63-5; 16141-96-1; and 14051-87-7.

The following are examples of peptides which are included within the generic formula L and which are identified by their Chemical Abstracts Registry Numbers: 104523-48-0; 104523-47-9; 104523-44-6; and 104523-43-5.

Nowhere do these structures in Chemical Abstracts teach or suggest the cyclic tripeptidic moieties of the compounds of the present invention.

SUMMARY OF THE INVENTION

The present invention particularly provides:

A compound of formula II wherein E_{10}-F_{11} is absent or a divalent moiety of the formula L_1, L_2, L_3, L_4 or L_5, or a monovalent moiety of the formula L_6, L_7 or L_8; wherein G_{12} is absent or a divalent moiety of the formula L_9;

wherein H_{13} is

a) -O-R_5, or

b) -N(R_1)(R_5);

wherein Q_1 is

a) -CH_2-,

b) -CH(OH),

c) -O-, or

d) -S-;

wherein M_1 is

a) -C(O)-, or

b) -CH_2-;

wherein V_1 is

a) -O-, or

b) -N(R_1)-;

wherein X_1 is

a) -Het,

b) $-N(R_1)-$, or

c) -S-;

wherein Y_1 is

a) -OH, or

b) $-NH_2$;

wherein Z_1 is

a) $-C(O)-(CH_2)_p-$, or

b) $-(CH_2)_p-C(O)-$;

wherein P_1 is

a) $-N_3$,

b) -CN,

c) C_1-C_6alkyl,

d) C_1-C_6cycloalkyl,

e) aryl, or

f) -Het;

wherein m is one or two;

wherein n is one to five, inclusive;

wherein p is zero to five, inclusive;

wherein aryl is phenyl or naphthyl, optionally substituted by 0 to 3 of the following:

(a) C_1-C_5alkyl,

(b) hydroxy,

(c) hydroxy(C_1-C_5alkyl),

(d) C_1-C_5alkoxy,

(e) amino,

(f) amino(C_1-C_5alkyl),

(g) halogen,

(h) -CHO,

(i) $-CO_2H$,

(j) $-CO_2-(C_1-C_5alkyl)$,

(k) $-CONH_2$,

(l) $-CONH(C_1-C_5alkyl)$,

(m) nitro,

(n) mercapto,

(o) mercapto(C_1-C_5alkyl),

(p) -SO_2H,

(q) -SO_2NH_2, or

(r) -CN;

wherein -Het is a 5- or 6-membered saturated or unsaturated ring containing from 1 to 3 heteroatoms (nitrogen, oxygen, or sulfur), and including any bicyclic group in which any of the above heterocyclic rings is fused to a benzene ring or another heterocycle, and if chemically feasible, the nitrogen and sulfur atoms may be in the oxidized forms; and optionally substituted by 0 to 3 of the following:

(a) C_1-C_5alkyl,

(b) hydroxy,

(c) hydroxy(C_1-C_5alkyl),

(d) C_1-C_5alkoxy,

(e) amino,

(f) amino(C_1-C_5alkyl),

(g) halogen,

(h) -CHO,

(i) -CO_2H,

(j) -CO_2-(C_1-C_5alkyl),

(k) -$CONH_2$,

(l) -CONH(C_1-C_5alkyl),

(m) nitro,

(n) mercapto,

(o) mercapto(C_1-C_5alkyl),

(p) -SO_2H,

(q) -SO_2NH_2, or

(r) -CN;

wherein R_1 is

(a) hydrogen, or

(b) C_1-C_5alkyl;

wherein R_2 is

(a) hydrogen,

(b) C_1-C_8alkyl,

(c) $-(CH_2)_p$-aryl,

(d) $-(CH_2)_p$-Het,

(e) $-(CH_2)_p-CO_2H$,

(f) $-(CH_2)_p-NH_2$,

(g) $-(CH_2)_p-CH(NH_2)(CO_2H)$,

(h) C_3-C_7cycloalkyl, or

(i) 1- or 2-adamantyl;

wherein R_3 is

(a) hydrogen,

(b) C_1-C_5alkyl,

(c) aryl,

(d) C_3-C_7cycloalkyl,

(e) -Het,

(f) C_1-C_3alkoxy, or

(g) C_1-C_3alkylthio;

wherein R_4 is

(a) hydrogen,

(b) C_1-C_8alkyl,

(c) C_3-C_7cycloalkyl, or

(d) $-CH(R_1)(R_6)$;

wherein R_5 is

(a) hydrogen,

(b) C_1-C_{10}alkyl.

(c) -(CH$_2$)p-alk

(d) $-(CH_2)_p$-het,

(e) $-(CH_2)_p$-cycloalkyl,

(f) $-(CH_2)_p-CH(NH_2)(CO_2H)$, or

(g) $-(CH_2)_n-R_7$;

wherein R_6 is

(a) hydrogen,

(b) hydroxy,

(c) C_1-C_5alkyl,

(d) C_3-C_7cycloalkyl,

(e) aryl,

(f) -Het,

(g) C_1-C_3alkoxy, or

(h) C_1-C_3alkylthio;

wherein R_7 is

(a) hydroxy,

(b) amino,

(c) $-CO_2H$,

(d) $-SO_2H$,

(e) $-SO_2NH_2$,

(f) guadinyl, or

(g) a polyhydroxylated-substituted-alkyl moiety;

or a carboxy-, amino- or other reactive group protected form thereof;

or a pharmaceutically acceptable acid or base addition salt thereof; provided that:

1) when E_{10}-F_{11} is the moiety of formula L_6, L_7 or L_8, then G_{12} and H_{13} are absent; and

2) when X_1 is -Het, it is bonded to $-(CH_2)_n$ of formula II as chemically feasible and is bonded to Z_1 of formula II by a heteroatom of -Het as chemically feasible.

These compounds are shown in relation to the human renin substrate as follows:

	6	7	8	9	10	11	12	13	
	-His	Pro	Phe	His	Leu	Val	Ile	His-	
X	A_6	B_7	C_8	D_9	E_{10}	F_{11}	G_{12}	H_{13}	I_{14} Z,

The present invention provides peptide inhibitors of renin and retroviral proteases which contain a novel cyclic tripeptide at the N-terminus of the peptide. Some of these compounds also have a non-cleavable transition state insert corresponding to the 10,11-position of the renin substrate (angiotensinogen).

By "renin inhibitory peptide" is meant a compound capable of inhibiting the renin enzyme in mammalian metabolism and having three or more amino acid residues linked by peptidic or pseudo-peptidic bonds.

By "a non-cleavable transition state insert" is meant a transition state insert which is not cleavable by a hydrolytic enzyme in mammalian metabolism. A variety of such transition state inserts, corresponding to the 10,11-position of the renin substrate, are known in the art, including those disclosed in the following references, which are hereby

incorporated by reference:

U.S. patent 4,424,207 (Szelke); European patent 104041A (Szelke); European patent application 144,290A (Ciba Geigy AG); European patent 0,156,322 (Merck); European patent 161-588A (Merck); European patent 0,172,347 (Abbott); European patent 172-346-A (Abbott); European patent 156-318 (Merck); European patent 157-409 (Merck); European patent 152-255 (Sankyo); and U.S. patent 4,548,926 (Sankyo); and

PCT International Application, Publication Number WO 87/05302, published 11 September 1987; PCT International Application, Publication Number WO 87/05909, published 8 October 1987; European application 0218688, published 22 April 1987; PCT International Application, Publication Number WO 88/04664, published 30 June 1988; and European application 0173481, published 5 March 1986; and

A. Spaltenstein, P. Carpino, F. Miyake and P.B. Hyskins, Tetrahedron Letters, 27:2095 (1986); D.H. Rich and M.S. Bernatowicz, J. Med. Chem., 25:791 (1982); Roger, J. Med. Chem., 28:1062 (1985); D.M. Glick, et al., Biochemistry, 21:3746 (1982); D.H. Rich, Biochemistry, 24:3165 (1985); R.L. Johnson, J. Med. Chem., 25:605 (1982); R.L. Johnson and K. Verschovor, J. Med. Chem., 26:1457 (1983); R.L. Johnson, J. Med. Chem., 27:1351 (1984); P.A. Bartlett, et al., J. Am. Chem. Soc., 106:4282 (1984); and Peptides: Synthesis, Structure and Function (V.J. Hruby; D.H. Rich, eds.) Proc. 8th American Peptide Sym., Pierce Chemical Company, Rockford, IL, pp. 511-20; 587-590 (1983).

By "derivatives" of amino acids is meant the well known amino acid derivatives commonly employed in renin inhibitors as set forth in the references above.

By "polyhydroxy-substituted-alkyl moiety" is meant that definition given on page 7 of PCT International Application PCT/US89/00247, Publication Number WO89/07109, published 10 August 1989, which is hereby incorporated by reference. An example of such a moiety is cyclohexylalaninol.

Examples of the peptides of the present invention are represented by formula II. In formula II, the non-cleavable transition state insert, corresponding to the 10,11-position of the renin substrate, is designated E_{10}-F_{11}.

The present invention provides novel peptides having a novel cyclic tripeptide at the N-terminus of the peptide. This new cyclic 14- to 19-membered ring spans the histidine residue at the P-2 site to the proline residue at the P-4 site of the angiotensinogen template. Peptides of the present invention having a cyclic 14-membered ring

tripeptide at the N-terminus are preferred. Some of the peptides of the present invention also have a non-cleavable transition state insert spanning the P_1-P_1' site of the angiotensinogen template.

In general, conformationally restricted peptides, such as cyclic peptides, have a number of advantages over linear peptides. In freezing out a large number of degrees of freedom of corresponding linear peptides, the pre-organized cyclic peptide can possess enhanced binding affinity to the target receptor. Cyclic peptides usually have increased resistance to proteolytic degradation relative to their linear counterparts. The large number of conformers of linear peptides which are available to proteases are essentially excluded by corresponding cyclic peptides. The very limited number of conformers of cyclic peptides do not present the proteases with optimal binding configurations for enzyme-catalyzed peptidic bond cleavage. Additionally, cyclic peptides are believed to exhibit much improved oral bioavailability over the congeneric linear peptides.

The peptides of the present invention are cyclic peptides and, as such, are believed to possess the advantages stated above for cyclic peptides in general. For example, cyclo-(Pro-Ala-N-MeHis)-NHMe has demonstrated increased oral bioavailability. In addition peptides cyclo-(Pro-Ala-N-MeHis)-NHMe and cyclo-(Pro-Phe-N-MeHis)-LVA-Ile-Amp are very highly water soluble.

In the text below, a shorthand notation has been used in some instances to refer to the cyclic tripeptide moieties of the compounds of the present invention. In this notation, the commonly used abbreviations of the three amino acids of the tripeptide are in parentheses with the word "cyclo" immediately preceding the parentheses. For example, by "cyclo-(Pro-Phe-His)" is meant the cyclic tripeptide moiety composed of the three amino acids Pro, Phe and His.

As is apparent to those of ordinary skill in the art, the renin inhibitory peptides of the present invention can occur in several diastereomeric forms, depending on the configuration around the asymmetric carbon atoms. All such diastereomeric forms are included within the scope of the present invention. Preferably, the stereochemistry of the amino acids corresponds to that of the naturally-occurring amino acids.

Renin inhibitory peptides commonly have protecting groups at the C-terminus. These protecting groups are known in the polypeptide art. Examples of these protecting groups are given below. Any of these protecting groups are suitable for the renin inhibitory peptides of the present invention.

Examples of pharmaceutically acceptable acid addition salts include: acetate, adipate, alginate, aspartate, benzoate, benzenesulfonate, bisulfate, butyrate, citrate, camphorate, camphorsulfonate, cyclopentanepropionate, digluconate, dodecylsulfate, ethane-sulfonate, fumarate, glucoheptanoate, glycerophosphate, hemisulfate, heptanoate, hexanoate, hydrochloride, hydrobromide, hydroiodide, 2-hydroxyethanesulfonate, lactate, maleate, methanesulfonate, 2-naphthalenesulfonate, nicotinate, oxalate, palmoate, pectinate, persulfate, 3-phenylpropionate, picrate, pivalate, propionate, succinate, tartrate, thiocyanate, tosylate, and undecanoate.

The carbon atom content of various hydrocarbon-containing moieties is indicated by a prefix designating the minimum and maximum number of carbon atoms in the moiety, i.e., the prefix (C_i-C_j) indicates a moiety of the integer "i" to the integer "j" carbon atoms, inclusive. Thus (C_1-C_4)alkyl refers to alkyl of one to 4 carbon atoms, inclusive, or methyl, ethyl, propyl, butyl, and isomeric forms thereof. C_4-C_7cyclic amino indicates a monocyclic group containing one nitrogen and 4 to 7 carbon atoms.

Examples of (C_3-C_{10})cycloalkyl, which include alkyl-substituted cycloalkyl containing a total of up to 10 total carbon atoms, are cyclopropyl, 2-methylcyclopropyl, 2,2-dimethylcyclopropyl, 2,3-diethylcyclopropyl, 2-butylcyclopropyl, cyclobutyl, 2-methylcyclobutyl, 3-propylcyclobutyl, cyclopentyl, 2,2-dimethylcyclopentyl, cyclohexyl, cycloheptyl, cyclooctyl, cyclononyl, cyclodecyl and isomeric forms thereof.

Examples of aryl include phenyl, naphthyl, (o-, m-, or p-)tolyl, (o-, m-, or p-)ethylphenyl, 2-ethyl-tolyl, 4-ethyl-o-tolyl, 5-ethyl-m-tolyl, (o-, m-, or p-)propylphenyl, 2-propyl-(o-, m-, or p-)tolyl, 4-isopropyl-2,6-xylyl, 3-propyl-4-ethylphenyl, (2,3,4-2,3,6-, or 2,4,5-)trimethylphenyl, (o-, m-, or p-)fluorophenyl, (o-, m-, or p-trifluoro-methyl)phenyl, 4-fluoro-2,5-xylyl, (2,4-, 2,5-, 2,6-, 3,4-, or 3,5-)difluorophenyl, (o-, m-, or p-)chlorophenyl, 2-chloro-p-tolyl, (3-, 4-, 5- or 6-)chloro-o-tolyl, 4-chloro-2-propylphenyl, 2-isopropyl-4-chlorophenyl, 4-chloro-3-fluorophenyl, (3- or 4-)chloro-2-fluorophenyl, (o-, m-, or p-)trifluoro-methylphenyl, (o-, m-, or p-)ethoxyphenyl, (4- or 5-)chloro-2-methoxy-phenyl, and 2,4-dichloro(5- or 6-)methylphenyl, and the like.

Examples of -Het include: 2-, 3-, or 4-pyridyl, imidazolyl, indolyl, N^{im}-formyl-indolyl, N^{im}-C_1-C_5alkyl-C(0)-indolyl, 1,2,4-triazolyl, 2-, 4-, or 5-pyrimidinyl, 2- or 3-thienyl, piperidinyl, pyrryl, pyrrolinyl, pyrrolidinyl, pyrazolyl, pyrazolinyl, pyrazolidinyl, imidazolinyl, imidazolidinyl, pyrazinyl, piperazinyl, pyridazinyl, oxazolyl, oxazolidinyl, isoxazolyl, isoxazolidinyl, morpholinyl, thiazolyl, thiazolidinyl, isothiazolyl,

isothiazolidinyl, quinolinyl, isoquinolinyl, benzimidazolyl, benzothiazolyl, benzoxazolyl, furyl, thienyl, and benzothienyl. Each of these moieties may be substituted as noted above.

By "chemically feasible" is meant that, given the knowledge available in the field of organic chemistry, it is possible for one of ordinary skill in the art to carry out the proposed chemical reaction, procedure or modification, and that what is "chemically feasible" would be readily apparent to one of ordinary skill in the art of organic chemistry. For example, as would be generally recognized by those skilled in the art of organic chemistry, a heterocycle as defined herein for -Het would not be bonded through oxygen or sulfur or through nitrogen which is within a ring and part of a double bond.

Halo is halogen (fluoro, chloro, bromo, or iodo) or trifluoromethyl.

Examples of pharmaceutically acceptable cations include: pharmacologically acceptable metal cations, ammonium, amine cations, or quaternary ammonium cations. Especially preferred metal cations are those derived from the alkali metals, e.g., lithium, sodium, and potassium, and from the alkaline earth metals, e.g., magnesium and calcium, although cationic forms of other metals, e.g., aluminum, zinc, and iron are also within the scope of this invention. Pharmacologically acceptable amine cations are those derived from primary, secondary, or tertiary amines.

The novel peptides herein contain both natural and synthetic amino acid residues. These residues are depicted using standard amino acid abbreviations (see, e.g., IUPAC-IUB Joint Commission on Biochemical Nomenclature (JCBN), "Nomenclature and Symbolism for Amino Acids and Peptides," Eur. J. Biochem. 138:9-37 (1984) unless otherwise indicated.

The peptides of this invention are useful for treating any medical condition for which it is beneficial to reduce the levels of active renin. Examples of such conditions include renin-dependent hypertension, hypertension, hypertension under treatment with another antihypertensive and/or a diuretic agent, congestive heart failure, renin-dependent hyperaldosterism, angina, post-myocardial infarction, other renin-dependent cardiovascular disorders and ocular disorders. The renin-angiotension system may play a role in maintenance of intracellular homeostasis: see Clinical and Experimental Hypertension, 86, 1739-1742 (1984) at page 1740 under Discussion.

The peptides of the present invention are preferably orally administered to

humans to effect renin inhibition for the purpose of favorably affecting blood pressure. For this purpose, the compounds are administered from 0.1 mg to 100 mg per kg per dose, administered from 1 to 4 times daily. Equivalent dosages for other routes of administration are also employed. For example, renin-associated hypertension and hyperaldosteronism are effectively treated by the administration of from 0.5 to 50 milligrams of the compound per kilogram of body weight per day. The exact dose depends on the age, weight, and condition of the patient and on the frequency and route of administration. Such variations are within the skill of the practitioner or can readily be determined.

The peptides of the present invention to effect renin inhibition may be in the form of pharmaceutically acceptable salts both those which can be produced from the free bases by methods well known in the art and those with which acids have pharmacologically acceptable conjugate bases.

Conventional forms and means for administering renin-inhibiting compounds may be employed and are described, e.g., in U.S. Patent No. 4,424,207 which is incorporated by reference herein. Likewise, the amounts disclosed in the U.S. Patent No. 4,424-,207 are examples applicable to the compounds of the present invention.

The peptides of the present invention to effect renin inhibition are preferably orally administered in the form of pharmacologically acceptable acid addition salts. Preferred pharmacologically acceptable salts for oral administration include the citrate and aspartate salts, although any pharmacologically acceptable salt is useful in this invention, including those listed above. These salts may be in hydrated or solvated form.

For renin inhibition, the peptides of the present invention may be administered topically, parenterally, by inhalation spray, or rectally in dosage unit formulations containing conventional non-toxic pharmaceutically acceptable carriers, adjuvants and vehicles. The term parenteral as used herein includes subcutaneous injections, intravenous, intramuscular, intrasternal injection or infusion techniques. In addition to the treatment of warm-blooded animals such as mice, rats, horses, dogs, cats, etc., the compounds of the invention are effective in the treatment of humans.

The pharmaceutical compositions of the peptides of the present invention for renin inhibition may be in the form of a sterile injectable preparation, for example as a sterile injectable aqueous or oleagenous suspension. This suspension may be formulated

according to the known art using suitable dispersing or wetting agents and suspending agents. The sterile injectable preparation may also be a sterile injectable solution or suspension in a non-toxic parenterally-acceptable diluent or solvent, for example as a solution in 1,3-butanediol. Among the acceptable vehicles and solvents that may be employed are water, Ringer's solution and isotonic sodium chloride solution. In addition, sterile, fixed oils are conventionally employed as a solvent or suspending medium. For this purpose any bland fixed oil may be employed including synthetic mono- or digly-cerides. In addition, fatty acids such as oleic acid find use in the preparation of injectables.

The peptides of this invention may also be administered in the form of suppositories for rectal administration of the drug. These compositions can be prepared by mixing the drug with a suitable non-irritating excipient which is solid at ordinary temperatures but liquid at the rectal temperature and will therefore melt in the rectum to release the drug. Such materials are cocoa butter and polyethylene glycols.

The peptides of this invention may be administered in combination with other agents used in antihypertensive therapy such as diuretics, α and/or ß-adrenergic blocking agents, CNS-acting agents, adrenergic neuron blocking agents, vasodilators, angiotensin I converting enzyme inhibitors, and the like as described for example in published European patent application 156,318.

The present invention is also directed to combinations of the novel renin-inhibitory peptides of Formula I with one or more antihypertensive agents selected from the group consisting of diuretics, α and/or ß-adrenergic blocking agents, CNS-acting agents, adrenergic neuron blocking agents, vasodilators, angiotensin I converting enzyme inhibitors, and other antihypertensive agents.

For example, the compounds of this invention can be given in combination with such compounds or salt or other derivative forms thereof as:

Diuretics: acetazolamide; amiloride; bendroflumethiazide; benzthia-zide; bumetanide; chlorothiazide; chlorthalidone; cyclothiazide; ethacrynic acid; furosemide; hydrochloro-thiazide; hydroflumethiazide; indacrinone (racemic mixture, or as either the (+) or (-) enantiomer alone, or a manipulated ratio, e.g., 9:1 of said enantiomers, respectively); metolazone; methyclothiazide; muzolimine; polythiazide; quinethazone; sodium ethacrynate; sodium nitroprusside; spironolactone; ticrynaten; trimaterene; trichlorme-thiazide;

α-Adrenergic Blocking Agents: dibenamine; phentolamine; phenoxybenzamine; prazosin; tolazoline;

ß-Adrenergic Blocking Agents: atenolol; metoprolol; nadolol; propranolol; timolol; ((±)-2-[3-(tert-butylamino)-2-hydroxypropoxy]-2-furananilide) (ancarolol);

(2-acetyl-7-(2-hydroxy-3-isopropylaminopropoxy)benzofuran HCl)(befunolol);

((±)-1-(isopropylamino)-3-(p-(2-cyclopropylmethoxyethyl)-phenoxy)-2-propranol HCl) (betaxolol);

(1-[(3,4-dimethoxyphenethyl)amino]-3-(m-tolyloxy)-2-propanol HCl)(bevantolol);

(((±)-1-(4-((2-isopropoxyethoxy)methyl)phenoxy)-3-isopropylamino-2-propanol)fumarate) (bisoprolol);

(4-(2-hydroxy-3-[4-(phenoxymethyl)-piperidino]-propoxy)-indole);

(carbazolyl-4-oxy-5,2-(2-methoxyphenoxy)-ethylamino-2-propanol);

(1-((1,1-dimethylethyl)amino)-3-((2-methyl 'H-indol-4-yl)oxy)-2-pro-panol benzoate) (bopindolol);

(1-(2-exobicyclo[2.2.1]-hept-2-ylphenoxy)-3-[(1-methylethyl)-amino]-2-propanol HCl) (bornaprolol);

(o-[2-hydroxy-3-[(2-indol-3-yl-1,1-dimethylethyl)-amino]propoxy]benzonitrile HCl) (bucindolol);

(α-[(tert.butylamino)methyl]-7-ethyl-2-benzofuranmethanol) (bufur-alol);

(3-[3-acetyl-4-[3-(tert.butylamino)-2-hydroxypropyl]-phenyl]-1,1-diethylurea HCl) (celiprolol);

((±)-2-[2-[3-[(1,1-dimethylethyl)amino]-2-hydroxypropoxy]phenoxy]-N-methylacetamide HCl) (cetamolol);

(2-benzimidazolyl-phenyl(2-isopropylaminopropanol));

((±)-3'-acetyl-4'-(2-hydroxy-3-isopropylaminopropoxy)-acetanilide HCl) (diacetolol);

(methyl-4-[2-hydroxy-3-[(1-methylethyl)aminopropoxyl]]-benzene-propanoate HCl) (esmolol);

(erythro-DL-1-(7-methylindan-4-yloxy)-3-isopropylaminobutan-2-ol);

(1-(tert.butylamino)-3-[0-(2-propynyloxy)phenoxy]-2-propanol (pargo-lol);

(1-(tert.butylamino)-3-[o-(6-hydrazino-3-pyridazinyl)phenoxy]-2-propanol diHCl) (prizidilol);

((-)-2-hydroxy-5-[(R)-1-hydroxy-2-[(R)-(1-methyl-3-phenylpropyl)-amino]ethyl]benzamide);

(4-hydroxy-9-[2-hydroxy-3-(isopropylamino)-propoxy]-7-methyl-5H-furo[3,2-g][1]-benzopyran-5-one) (iprocrolol);

((-)-5-(tert.butylamino)-2-hydroxypropoxy]-3,4-dihydro-1-(2H)-naphthalenone HCl) (levobunolol);

(4-(2-hydroxy-3-isopropylamino-propoxy)-1,2-benzisothiazole HCl);

(4-[3-(tert.butylamino)-2-hydroxypropoxy]-N-methylisocarbostyril HCl);

((±)-N-2-[4-(2-hydroxy-3-isopropylaminopropoxy)phenyl]ethyl-N'-isopropylurea) (pafenolol);

(3-[[(2-trifluoroacetamido)ethyl]amino]-1-phenoxypropan-2-ol);

(N-(3-(o-chlorophenoxy)-2-hydroxypropyl)-N'-(4'-chloro-2,3-dihydro-3-oxo-5-pyridazinyl)ethylenediamine);

((±)-N-[3-acetyl-4-[2-hydroxy-3-[(1-methylethyl)amino]propoxyphenyl]-butanamide) (acebutolol);

((±)-4'-[3-(tert-butylamino)-2-hydroxypropoxy]spiro[cyclohexane-1,2'-indan]-1'-one) (spirendolol);

(7-[3-[[2-hydroxy-3-[(2-methylindol-4-yl)oxylpropyl]amino]butyl]thio-phylline) (teoprolol);

((±)-1-tert.butylamino-3-(thiochroman-8-yloxy)-2-propanol) (tertato-lol);

((±)-1-tert.butylamino-3-(2,3-xylyloxy)-2-propanol HCl) (xibenolol);

(8-[3-(tert.butylamino)-2-hydroxypropoxy]-5-methylcoumarin) (bucumo-lol);

(2-(3-(tert.butylamino)-2-hydroxy-propoxy)benzonitrile HCl) (bunitro-lol);

((±)-2'-[3-(tert-butylamino)-2-hydroxypropoxy-5'-fluorobutyrophenone) (butofilolol);

(1-(carbazol-4-yloxy)-3-(isopropylamino)-2-propanol) (carazolol);

(5-(3-tert.butylamino-2-hydroxy)propoxy-3,4-dihydrocarbotyril HCl) (carteolol);

(1-(tert.butylamino)-3-(2,5-dichlorophenoxy)-2-propanol) (clorano-lol);

(1-(inden-4(or 7)-yloxy)-3-(isopropylamino)-2-propanol HCl) (indeno-lol);

(1-isopropylamino-3-[(2-methylindol-4-yl)oxy]-2-propanol) (mepindo-lol);

(1-(4-acetoxy-2,3,5-trimethylphenoxy)-3-isopropylaminopropan-2-ol) (metipranolol);

(1-(isopropylamino)-3-(o-methoxyphenoxy)-3-[(1-methylethyl)amino]-2-propanol) (moprolol);

((1-tert.butylamino)-3-[(5,6,7,8-tetrahydro-cis-6,7-dihydroxy-1-naphthyl)oxy]-2-propanol) (nadolol);

((S)-1-(2-cyclopentylphenoxy)-3-[(1,1-dimethylethyl)amino]-2-propanol sulfate (2:1))

(penbutolol);

(4'-[1-hydroxy-2-(amino)ethyl]methanesulfonanilide) (sotalol);

(2-methyl-3-[4-(2-hydroxy-3-tert.butylaminopropoxy)phenyl]-7-methoxy-isoquinolin-1-(2H)-one);

(1-(4-(2-(4-fluorophenyloxy)ethoxy)phenoxy)-3-isopropylamino-2-propanol HCl);

((-)-p-[3-[(3,4-dimethoxyphenethyl)amino]-2-hydroxypropoxy]-ß-methyl-cinnamonitrile) (pacrinolol);

((±)-2-(3'-tert.butylamino-2'-hydroxypropylthio)-4-(5'-carbamoyl-2'-thienyl)thiazole HCl) (arotinolol);

((±)-1-[p-[2-(cyclopropylmethoxy)ethoxy]phenoxy]-3-(isopropylamino)-2-propanol) (cicloprolol);

((±)-1-[(3-chloro-2-methylindol-4-yl)oxy]-3-[(2-phenoxyethyl)amino]-2-propanol) (indopanolol);

((±)-6-[[2-[[3-(p-butoxyphenoxy)-2-hydroxypropyl]amino]ethyl]amino]-1,3-dimethyluracil) (pirepolol);

(4-(cyclohexylamino)-1-(1-naphtholenyloxy)-2-butanol);

(1-phenyl-3-[2-[3-(2-cyanophenoxy)-2-hydroxypropyl]aminoethyl]hydan-toin HCl);

(3,4-dihydro-8-(2-hydroxy-3-isopropylaminopropoxy)-3-nitroxy-2H-1-benzopyran) (nipradolol);

Angiotensin I Converting Enzyme Inhibitors:

1-(3-mercapto-2-methyl-1-oxopropyl)-L-proline (captopril);

(1-(4-ethoxycarbonyl-2,4(R,R)-dimethylbutanoyl)indoline-2(S)-carboxylic acid);

(2-[2-[(1-(ethoxycarbonyl)-3-phenyl-propyl]amino]-1-oxopropyl]-1,2,3,4-tetrahydro-3-isoquinoline carboxylic acid);

((S)-1-[2-[(1-(ethoxycarbonyl)-3-phenylpropyl]amino]-1-oxopropyl]octahydro-1H-indole-2-carboxylic acid HCl);

(N-cyclopentyl-N-(3-(2,2-dimethyl-1-oxopropyl)thiol-2-methyl-1-oxo-propyl)glycine) (pivalopril);

((2R,4R)-2-(2-hydroxyphenyl)-3-(3-mercaptopropionyl)-4-thiazolidine-carboxylic acid);

(1-(N-[1(S)-ethoxycarbonyl-3-phenylpropyl]-(S)-alanyl)-cis,syn-octa-hydroindol-2(S)-carboxylic acid HCl);

((-)-(S)-1-[(S)-3-mercapto-2-methyl-1-oxopropyl]indoline-2-carboxylic acid);

([1(S),4S]-1-[3-(benzoylthio)-2-methyl-1-oxopropyl]-4-phenylthio-L-proline;

(3-([1-ethoxycarbonyl-3-phenyl-(1S)-propyl]amino)-2,3,4,5-tetrahydro-2-oxo-1-(3S)-benzazepine-1-acetic acid HCl);

(N-(2-benzyl-3-mercaptopropanoyl)-S-ethyl-L-cysteine) and the S-methyl analogue;

(N-(1(S)-ethoxycarbonyl-3-phenylpropyl)-L-alanyl-L-proline maleate) (enalapril);

N-[1-(S)-carboxy-3-phenylpropyl]-L-alanyl-1-proline;

N^2-[1-(S)-carboxy-3-phenylpropyl]-L-lysyl-L-proline (lysinopril);

Other Antihypertensive Agents: aminophylline; cryptenamine acetates and tannates; deserpidine; meremethoxylline procaine; pargyline; tri-methaphan camsylate; and the like, as well as admixtures and combinations thereof.

Typically, the individual daily dosages for these combinations can range from about one-fifth of the minimally recommended clinical dosages to the maximum recommended levels for the entities when they are given singly. Coadministration is most readily accomplished by combining the active ingredients into a suitable unit dosage form containing the proper dosages of each. Other methods of coadministration are, of course, possible.

Thus, the novel peptides of the present invention possess an excellent degree of activity in treating renin-associated hypertension and hyperaldosteronism.

Renin inhibitors have also been disclosed to control the rise in intraocular pressure associated with the use of steroidal anti-inflammatory drugs as described in International Application PCT/ US86/02291 (International Publication Number WO 87/02581 dated 7 May 1987).

The peptides of the present invention are also useful as novel human retroviral protease inhibitory peptide analogs. Therefore, the peptides of the present invention inhibit retroviral proteases and thus inhibit the replication of the virus. They are useful for treating human patients infected with a human retrovirus, such as human immunodeficiency virus (strains of HIV-1 or HIV-2) or human T-cell leukemia viruses (HTLV-I or HTLV-II) which results in acquired immunodeficiency syndrome (AIDS) and/or related diseases.

The capsid and replicative enzymes (i.e. protease, reverse transcriptase, integrase) of retroviruses are translated from the viral gag and pol genes as polyproteins that are further processed by the viral protease (PR) to the mature proteins found in the viral capsid and necessary for viral functions and replication. If the PR is absent or nonfunctional, the virus cannot replicate. The retroviral PR, such as HIV-1 PR, has been found

to be an aspartic protease with active site characteristics similar to those exhibited by the more complex aspartic protease, renin.

The term human retrovirus (HRV) includes human immunodeficiency virus type I, human immunodeficiency virus type II, or strains thereof, as well as human T cell leukemia virus 1 and 2 (HTLV-1 and HTLV-2) or strains apparent to one skilled in the art, which belong to the same or related viral families and which create similar physiological effects in humans as various human retroviruses.

Patients to be treated would be those individuals: 1) infected with one or more strains of a human retrovirus as determined by the presence of either measurable viral antibody or antigen in the serum and 2) in the case of HIV, having either a symptomatic AIDS defining infection such as i) disseminated histoplasmosis, ii) isopsoriasis, iii) bronchial and pulmonary candidiasis including pneumocystic pneumonia iv) non-Hodgkin's lymphoma or v) Kaposi's sarcoma and being less than sixty years old; or having an absolute CD4 lymphocyte count of less than 200/m^3 in the peripheral blood. Treatment would consist of maintaining an inhibitory level of the peptide used according to this invention in the patient at all times and would continue until the occurrence of a second symptomatic AIDS defining infection indicates alternate therapy is needed.

More specifically, an example of one such human retrovirus is the human immunodeficiency virus (HIV, also known as HTLV-III or LAV) which has been recognized as the causative agent in human acquired immunodeficiency disease syndrome (AIDS), P. Duesberg, Proc. Natl. Acad. Sci. USA, 86:755 (1989). HIV contains a retro viral encoded protease, HIV-I protease, that cleaves the fusion polypeptides into the functional proteins of the mature virus particle, E.P. Lillehoj, et al., J. Virology, 62:3053 (1988); C. Debuck, et al., Proc. Natl. Acad. Sci., 84:8903 (1987). This enzyme, HIV-I protease, has been classified as an aspartyl protease and has a demonstrated homology to other aspartyl proteases such as renin, L.H. Pearl, *et al.*, Nature 329:351 (1987); I. Katoh, *et al.*, Nature 329:654 (1987). Inhibition of HIV-I protease blocks the replication of HIV and thus is useful in the treatment of human AIDS, E.D. Clerq, J. Med. Chem. 29:1561 (1986). Inhibitors of HIV-I protease are useful in the treatment of AIDS.

Pepstatin A, a general inhibitor of aspartyl proteases, has been disclosed as an inhibitor of HIV-I protease, S. Seelmeier, *et al.*, Proc. Natl. Acad. Sci. USA, 85:6612 (1986). Other substrate derived inhibitors containing reduced bond isosteres or statine

at the scissle position have also been disclosed, M.L. Moore, *et al.*, Biochem. Biophys, Res. Commun. 159:420 (1989); S. Billich, *et al.*, J. Biol. Chem. 263:17905 (1988); Sandoz, D.E. 3812-576-A.

Thus, the peptides of the present invention are useful for treating diseases caused by retroviruses, such as human acquired immunodeficiency disease syndrome (AIDS), using dosages, forms and modes of administration equivalent to those described above for renin inhibition. Exact dosages, forms and mode of administration would be apparent to one of ordinary skill in the art such as a physician or pharmacologist.

The peptides of the present invention are also useful for treating non-human animals infected with a retrovirus, such as cats infected with feline leukemia virus. Other viruses that infect cats include, for example, feline infectious peritonitis virus, calicivirus, rabies virus, feline immunodeficiency virus, feline parvovirus (panleukopenia virus), and feline chlamydia. Exact dosages, forms and modes of administration of the peptides of the present invention to non-human animals would be apparent to one of ordinary skill in the art, such as a veterinarian.

The HIV protease inhibitor activity of the compounds of the present invention has been demonstrated in the biological test described below.

The HIV-1 protease has been expressed in E. coli, isolated, characterized and used to determine the inhibitory constants (K_i) of potential inhibitory compounds as follows:

The synthetic peptide H-Val-Ser-Gln-Asn-Tyr-Pro-Ile-Val-OH serves as the substrate for the measurement of HIV-1 protease activity. This peptide corresponds to the sequence from residue 128 to 135 in the HIV gag protein. Cleavage of the synthetic peptide, as well as the gag protein, takes place at the Tyr-Pro bond. HIV-1 protease activity is measured at 30°C in 50 mM sodium acetate, pH 5.5, containing 10% glycerol, 5% ethylene glycol, 0.1% Nonidet P-40 and 2.8 mM substrate in a total volume of 50 μl. After 30 minutes of incubation, 75 μl of 1% trifluoroacetic acid (TFA) is added and the reaction mixture subjected to HPLC analysis. HPLC is carried out with a Vydac C_{18} column (0.46 x 15 cm), eluting with a linear gradient of 0-30% acetonitrile over a period of 25 minutes at a flow rate of 1.0 ml/minute.

The K_i value or % inhibition of compounds of the present invention are given in Examples 3, 7 and 12 below.

The compounds of the present invention are prepared as depicted in the charts and

as described more fully in the Preparations and Examples. In these charts, the variables are as defined above.

Chart A

Chart A describes the preparation of the peptide of the present invention of the formula A-2. The compound of formula A-1, prepared as described in U.S. patent application, Serial No. 07/339,750, filed 18 April 1989, is reacted with bromoacetyl bromide which is commercially available to obtain the cyclic final peptide of formula A-2.

Chart B and Chart C (Scheme 1)

Chart B and Chart C (Scheme 1) describe the preparation of the peptides of the present invention of the formula B-5 and of the formula C-4, respectively. The compound of formula B-4 is obtained in a straightforward reaction sequence from the compound of formula B-1, using chemical reactants and procedures which are readily known and available to one of ordinary skill in the art. The compound of formula B-4 is deprotected and treated with bromoacetyl bromide to obtain the cyclic final peptide of formula B-5.

In Chart C (Scheme 1), the compound of formula C-3, is obtained in a straightforward reaction sequence from the compound of formula C-1, using chemical reactants and procedures which are readily known and available to one of ordinary skill in the art. The compound of formula C-3 is deprotected and treated with bromoacetyl bromide to obtain the cyclic final peptide of formula C-4.

Chart C (Scheme 2)

Chart C (Scheme 2) describes an alternative method for the preparation of the peptide of the present invention of formula C-4. The compound of formula C-3 is treated with tert-butyl bromoacetate to afford the imidazole-alkylated material of formula C-5. The Boc group on proline and the tert-butyl ester are removed with trifluoroacetic acid to obtain the compound of formula C-6. The resulting amino group is cyclized under high dilution with benzotriazol-1-yloxytris(dimethylamino)phosphonium hexafluorophosphate (BOP) reagent and 4-dimethylaminopyridine (DMAP) in dichloromethane to give the final cyclic peptide of formula C-4.

Chart D

Chart D describes the preparation of the peptide of the present invention of the formula D-7 having a non-methylated histidine residue. The compound of formula D-4

is obtained in a straightforward reaction sequence from the compound of formula D-1 using chemical reactants and procedures which are readily known and available to one of ordinary skill in the art. The compound of formula D-4 is treated with tert-butyl bromoacetate to afford the alkylated compound of formula D-5. The Boc group and the tert-butyl ester are removed with trifluoroacetic acid to obtain the compound of formula D-6. This compound is cyclized with BOP reagent and DMAP under high dilution to give the final cyclic peptide of formula D-7.

Chart E

Chart E describes the preparation of the peptide of the present invention of formula E-6 and of the intermediate compound of formula E-7.

Protected histidine derivative of formula E-1, prepared by methods known in the art, is first de-Boc'd, and the resulting free amine coupled with Boc-Pro-Phe-OH acid to yield the tripeptide derivative of formula E-2. Treatment of the compound of formula E-2 with 1-hydroxybenzotriazole monohydrate (HOBT) in methanol removes the tosyl group protection from the histidine imidazole ring to give the compound of formula E-3, which is then alkylated with tert-butyl bromoacetate to give the compound of formula E-4. Simultaneous removal of the Boc and tert-butyl ester groups of the compound of formula E-4 gives the compound of formula E-5. The cyclization of this compound is accomplished with BOP reagent and DMAP in dichloromethane under high dilution (ca. 0.0015 M) to provide the final macrocycle peptide of formula E-6. Finally, transfer hydrogenolysis of the benzyl ester in the compound of formula E-6 provides the acid of formula E-7, which is suitable for coupling to a variety of other amino acid residues to prepare other peptides of the present invention. BOP reagent is the preferred coupling reagent. Such coupling procedures are analogous to those described above and are readily known and available to one of ordinary skill in the art.

Chart F

Chart F describes the preparation of the peptide of the present invention of formula F-5. The tosyl protection of the compound of formula F-1, prepared as described in PCT International Application PCT/US89/00247, Publication Number WO89/ 07109 published 10 August 1989, is removed with 1-hydroxybenzotriazole monohydrate (HOBT) in methanol to give the compound of formula F-2. The free imidazole ring is alkylated to give the compound of formula F-3. Proteolysis of the acid labile groups of the compound of formula F-3 gives the compound of formula F-4. This

compound is cyclized using BOP reagent and DMAP in dichloromethane under high dilution (ca. 0.0015 M) to provide the final peptide of formula F-5.

Additional peptides of the present invention of formula II wherein the variables are as defined above may be prepared by procedures which are analogous to those described above and which are readily known and available to one of ordinary skill in the art of peptide synthesis.

Generally, the renin inhibiting polypeptides may be prepared by solution phase peptide synthetic procedures analogous to those described hereinafter or to those methods known in the art. Appropriate protecting groups, reagents, and solvents for the solution phase method can be found in "The Peptides: Analysis, Synthesis, and Biology," Vols. 1-5, eds. E. Gross and T. Meienhofer, Academic Press, NY, 1979-1983; "The Practice of Peptide Synthesis", M. Bodansky and A. Bodansky, Springer-Verlag, New York, 1984; "The Principles of Peptide Synthesis", M. Bodansky, Springer-Verlag, New York, 1984. Thus, for example, the carboxylic moiety of N^{α}-t-butyloxycarbonyl (Boc)-substituted amino acid derivatives having suitable side chain protecting groups, if necessary, may be condensed with the amino functionality of a suitably protected amino acid or peptide using a conventional coupling protocol such as dicyclohexylcarbodiimide (DCC) and HOBT or DEPC and triethylamine (Et_3N) in methylene chloride or dimethyl-formamide.

Following coupling reaction completion, the N^{α}-Boc moiety may be selectively removed with 50% trifluoroacetic acid with or without 2% anisole (v/v) in methylene chloride. Neutralization of the resultant trifluoroacetate salt may be accomplished with 10% diisopropylethylamine or sodium bicarbonate in methylene chloride.

Variations in the above description for starting materials, reactants, reaction conditions and required protecting groups to obtain other such N-alkylated compounds are known to an ordinarily skilled chemist or are readily available in the literature.

The compounds of the present invention may be in either free form or in protected form at one or more of the remaining (not previously protected) peptide, carboxyl, amino, hydroxy, or other reactive groups. The protecting groups may be any of those known in the polypeptide art. Examples of nitrogen and oxygen protection groups are set forth in T.W. Greene, Protecting Groups in Organic Synthesis, Wiley, New York, (1981); J.F.W. McOmie, ed. Protective Groups in Organic Chemistry, Plenum Press (1973); and J. Fuhrhop and G. Benzlin, Organic Synthesis, Verlag Chemie

(1983). Included among the nitrogen protective groups are t-butoxycarbonyl (Boc), benzyloxycarbonyl, acetyl, allyl, phthalyl, benzyl, benzoyl, trityl and the like.

The following compounds of the present invention are preferred: cyclo-(Pro-Phe-N-MeHis)-NHMeorL-Histidinamide,L-prolyl-L-phenylalanyl-1-(carboxymethyl)-N,Nα-dimeth-yl-, cyclic (3→1)- peptide;

cyclo-(Pro-Ala-His)-NHMe or L-Histidinamide, L-prolyl-L-alanyl-1-(carboxymethyl)-N-methyl-, cyclic (3→1)-peptide;

cyclo(Pro-Ala-His)-CVA-Ile-Amp; or L-Histidinamide, L-prolyl-L-alanyl-1-(carboxymethyl)-N-[1-(cyclohexylmethyl)-2-hydroxy-5-methyl-4-[[[2-methyl-1-[[(2-pyridinylmethyl)amino]carbonyl]butyl]amino]carbonyl]hexyl],cyclic (3→1)-peptide, [1S-[1R*,2R*,4R*(1R*,2R*)]]-; cyclo(Pro-Ala-His)-CVA-Mba; or L-Histidinamide, L-prolyl-L-alanyl-1-(carboxymethyl)-N-[1-cyclohexylmethyl)-2-hydroxy-5-methyl-4-[[[(2-methylbutyl)amino]carbonyl]hexyl], cyclic (3→1)-peptide, [1S-[1R*,2R*,4R*(2R*)]]-;

cyclo(Pro-Ala-His)-CVA-Ile-Amp→O; or L-Histidinamide, L-prolyl-L-alanyl-1-(carboxymethyl)-N-[1-cyclohexylmethyl)-2-hydroxy-5-methyl-4-[[[(2-methyl-1-[[(2-pyridinylmethyl, N-oxide)amino]carbonyl]butyl]amino]carbonyl]hexyl], cyclic (3→1)-peptide, [1S-[1R*,2R*, 4R*(1R*,2R*)]]-;

cyclo-(Pro-Phe-N-MeHis)-LVA-Ile-Amp or L-Histidinamide, L-prolyl-L-phenylalanyl-1-(carboxymethyl)-N-[2-hydroxy-5-methyl-1-(2-methylpropyl)-4-[[[2-methyl-1-[[(2-pyridinylmethyl)amino]carbonyl]butyl]amino]carbonyl]hexyl]-Nα-methyl-, cyclic (3→1)-peptide, [1S-[1R*,2R*,4R*(1R*,2R*)]]-;

cyclo-(Pro-Ala-N-MeHis)-NHMe or L-Histidinamide, L-prolyl-L-alanyl-1-(carboxymethyl)-N,Nα-dimethyl-, cyclic (3→1)-peptide;

cyclo-[Pro-Phe-(NMe)His]-CVG or L-Histidinamide, L-prolyl-L-phenylalanyl-1-(carboxymethyl)-N-[[1-(cyclohexylmethyl)-2,3-dihydroxy-5-methyl]hexyl]-Nα-methyl-, cyclic (3→1)-peptide, [1S-[1R*,2S*,3R*]]-;

cyclo(Pro-Phe-His)-cyclohexyl-alaninol or L-Histidinamide, L-prolyl-L-phenylalanyl-1-(carboxymethyl)-N-[(1-cyclohexylmethyl-2-hydroxy)ethyl],cyclic (3→1)-peptide, [1S]-;

cyclo-[Pro-Phe-His]-CVA-Mba or L-Histidinamide, L-prolyl-L-phenylalanyl-1-(carboxymethyl)-N-[1-(cyclohexylmethyl)-2-hydroxy-5-methyl-4-[[[(2-methylbutyl-)amino]carbonyl]hexyl], cyclic (3→1)-peptide, [1S-[1R*,2R*,4R*(2R*)]]-; or

cyclo-[Pro-Phe-His]-CVA-Ile-Amp or L-Histidinamide, L-prolyl-L-phenylalanyl-

1-(carboxymethyl)-N-[1-(cyclohexylmethyl)-2-hydroxy-5-methyl-4-[[[2-methyl-1-[[(2-pyridinylmethyl)amino]carbonyl]butyl]amino]carbonyl]hexyl],cyclic (3→1)-peptide, [1S-[1R*,2R*,4R*(1R*, 2R*)]]-.

The following compounds of the present invention are more preferred:

cyclo-(Pro-Phe-N-MeHis)-LVA-Ile-Amp or L-Histidinamide, L-prolyl-L-phenylalanyl-1-(carboxymethyl)-N-[2-hydroxy-5-methyl-1-(2-methylpropyl)-4-[[[2-methyl-1-[[(2-pyridinylmethyl)amino]carbonyl]butyl]amino]carbonyl]hexyl]-Nα-methyl-, cyclic (3→1)-peptide, [1S-[1R*,2R*,4R*(1R*,2R*)]]-;

cyclo-(Pro-Ala-N-MeHis)-NHMe or L-Histidinamide, L-prolyl-L-alanyl-1-(carboxymethyl)-N,Nα-dimethyl-, cyclic (3→1)-peptide;

cyclo-[Pro-Phe-(NMe)His]-CVG or L-Histidinamide, L-prolyl-L-phenylalanyl-1-(carboxymethyl)-N-[[1-(cyclohexylmethyl)-2,3-dihydroxy-5-methyl]hexyl]-Nα-methyl-, cyclic (3→1)-peptide, [1S-[1R*,2S*,3R*]]-; or

cyclo(Pro-Phe-His)-cyclohexyl-alaninol or L-Histidinamide, L-prolyl-L-phenylalanyl-1-(carboxymethyl)-N-[(1-cyclohexylmethyl-2-hydroxy)ethyl],cyclic (3→1)-peptide, [1S]-.

The following compounds of the present invention are most preferred:

cyclo-[Pro-Phe-His]-CVA-Mba or L-Histidinamide, L-prolyl-L-phenylalanyl-1-(carboxymethyl)-N-[1-(cyclohexylmethyl)-2-hydroxy-5-methyl-4-[[[(2-methylbutyl-)amino]carbonyl]hexyl], cyclic (3→1)-peptide, [1S-[1R*,2R*,4R*(2R*)]]-; or

cyclo-[Pro-Phe-His]-CVA-Ile-Amp or L-Histidinamide, L-prolyl-L-phenylalanyl-1-(carboxymethyl)-N-[1-(cyclohexylmethyl)-2-hydroxy-5-methyl-4-[[[2-methyl-1-[[(2-pyridinylmethyl)amino]carbonyl]butyl]amino]carbonyl]hexyl],cyclic (3→1)-peptide, [1S-[1R*,2R*,4R*(1R*, 2R*)]]-.

DESCRIPTION OF THE PREFERRED EMBODIMENTS

The following Preparations and Examples illustrate the present invention.

In the Preparations and Examples below and throughout this document:

^{1}H-NMR is nuclear magnetic resonance

Ala is alanine

Amp is 2-aminomethylpyridine

Amp→O is 2-aminomethylpyridine N-oxide

Boc is t-butoxycarbonyl

BOP reagent is benzotriazol-1-yloxytris(dimethylamino)phosphonium hexaflu-

orophosphate

Bz is benzyl

C is centigrade

Cbz is benzyloxycarbonyl

$CDCl_3$ is deuteriochloroform

Celite is a filter aid

CVA is ChaΨ[CH(OH)CH_2]Val

CVG is chaval glycol of formula XX

DCC is dicyclohexylcarbodiimide

DEPC is diethylphosphoryl cyanide

Et0Ac is ethyl acetate

g is grams

τ-Glu is τ-glutamic acid

His is histidine

N-MeHis is Nα-methyl histidine

HOBT is 1-hydroxybenzotriazole

HPLC is high performance liquid chromatography

Ile is isoleucine

IR is infrared spectra

LVA is LeuΨ(CH(0H)CH_2)Val with the S configuration at C4 (the hydroxyl-bearing carbon atom)

M or mol is mole

Mba is 2S-methylbutylamine

Me is methyl

ml is milliliter

MPLC is medium pressure liquid chromatography

MS is mass spectroscopy

Ph is phenyl

Phe is phenylalanine

Pro is proline

RIP means a compound having the formula H-Pro-His-Phe-His-Phe-Phe-Val-Tyr-Lys-0H.2($CH_3C(0)0H$).XH_20 which is a known renin-inhibiting peptide

TBAP is tetra-n-butylammonium phosphate

TEA is triethylamine

TFA is trifluoroacetic acid

THF is tetrahydrofuran

TLC is thin layer chromatography

Tos is p-toluenesulfonyl

TsOH is p-toluenesulfonic acid

Tyr is tyrosine

(OCH$_3$)Tyr is O-methyl tyrosine

ß-Val is ß-Valine.

The wedge-shape line indicates a bond which extends above the plane of the paper relative to the plane of the compound thereon.

The dotted line indicates a bond which extends below the plane of the paper relative to the plane of the compound thereon.

In the examples below, the activity of the compounds of the present invention (IC_{50}'s) are determined using the in vitro test described in U.S. patent application, Serial No. 07/147,073, filed 20 January 1988, and in published European patent application 0 173 481, published 5 March 1986, pages 103-105, which are hereby incorporated by reference.

In the examples below, HPLC is high pressure liquid chromatography and k' is the partition ratio obtained. The solvent system used is indicated in parentheses after the partition ratio. HPLC is performed on a Perkin-Elmer Series IV liquid chromatograph operating at 1.5 ml/min through a Brownlee RP-18 10 micron column, with UV monitoring by a Kratos Spectroflow 773 detector. A Perkin-Elmer LCI-100 integrator is used for peak data. Solvent A is methanol; solvent B is an aqueous phosphate buffer containing 5.22 g NaH_2PO_4 monohydrate and 0.76 ml of 85% phosphoric acid in 4L of Burdick & Jackson water; solvent C is acetonitrile containing 1 g/L of TBAP; solvent D, which also contains 1 g/l OF TBAP is 90% vol B and 10% acetonitrile; solvent E is 10% acetonitrile in water with 0.1% TFA; solvent F is 10% water in acetonitrile with 0.1% TFA.

Example 1 cyclo-(Pro-Phe-N-MeHis)-LVA-Ile-Amp or L-Histidinamide, L-prolyl-L-phenylalanyl-1-(carboxymethyl)-N-[2-hydroxy-5-methyl-1-(2-methylpropyl)-4-[[[2-methyl-1-[[(2-pyridinylmethyl)amino]-carbonyl]butyl]amino]carbonyl]hexyl]-Nα-methyl-, cyclic (3→1)-

peptide, [1S-[1R*,2R*,4R*(1R*,2R*)]]- (Formula A-2: refer to Chart A.)

To a cold (-20°C), stirred solution of 100 mg of H-Pro-Phe-N-MeHis-LVA-Ile-Amp of formula A-1 and 0.013 ml of pyridine in 1 ml of dichloromethane is slowly added 11.5 μl of bromoacetyl bromide. The reaction mixture is allowed to warm to room temperature, and after one hour it is partitioned between aqueous sodium bicarbonate and dichloromethane/methanol. The organic phase is dried (sodium sulfate) and then concentrated. The residue is chromatographed on silica gel with 3%-10% methanol in dichloromethane to give 44 mg of the title product. Physical characteristics are as follows:

FAB-HRMS: (m+H) = 870.5236.

IC_{50}: 1.6×10^{-8} M.

Preparation 1 Boc-N-MeHis(Ts)-NHMe (Formula B-1: refer to Chart B.)

To a stirred solution of 1.0 g of Boc-N-MeHis(Ts)-OH and 320 mg of methylamine hydrochloride in 10 ml of dichloromethane is added 1.5 ml of diisopropylethylamine, followed by 0.38 ml of diethylphosphoryl cyanide. The reaction mixture is stirred overnight, then flash chromatographed on silica gel with ethyl acetate to afford 657 mg of the title product. The proposed structure is supported by ^{1}H-NMR.

Preparation 2 Boc-Phe-N-MeHis(Ts)-NHMe (Formula B-2: refer to Chart B).

A solution of 330 mg of the title product of Preparation 1 in 1 ml of dichloromethane and 1 ml of trifluoroacetic acid is allowed to stir for 45 minutes. The mixture is then added to a solution of 1.5 g of sodium bicarbonate in 15 ml of water. The resulting mixture is extracted with several portions of dichloromethane. The combined organic phase is dried (magnesium sulfate) and then concentrated to give the free amine.

To a stirred solution of this residue and 308 mg of Boc-Phe-OH in 2.5 ml of dichloromethane is added 0.27 ml of diisopropylethylamine, followed by 310 mg of bis(2-oxo-3-oxazolidinyl)phosphinic chloride. The reaction mixture is stirred overnight, then flash chromatographed on silica gel with ethyl acetate to afford 360 mg of the title product. The proposed structure is supported by ^{1}H-NMR.

Preparation 3 Boc-Pro-Phe-N-MeHis(Ts)-NHMe (Formula B-3: refer to Chart B.)

A solution of 360 mg of the title product of Preparation 2 in 1 ml of dichloromethane and 1 ml of trifluoroacetic acid is allowed to stir for 45 minutes. The mixture

is then added to a solution of 1.5 g of sodium bicarbonate in 15 ml of water. The resulting mixture is extracted with several portions of dichloromethane. The combined organic phase is dried (magnesium sulfate) and then concentrated to give the free amine.

To a stirred solution of this residue and 153 mg of Boc-Pro-OH in 4 ml of dichloromethane is added 160 μl of diisopropylethylamine, followed by 110 μl of diethylphosphoryl cyanide. The reaction mixture is stirred overnight, then flash chromatographed on silica gel with 4% methanol in ethyl acetate to afford 388 mg of the title product. The proposed structure is supported by ^{1}H-NMR.

Preparation 4 Boc-Pro-Phe-N-MeHis-NHMe (Formula B-4: refer to chart B.)

A solution of 388 mg of the title product of Preparation 3 and 400 mg of 1-hydroxybenzotriazole monohydrate in 2 ml of methanol is allowed to stand overnight and is then concentrated under reduced pressure. Chromatography of the residue on silica gel with 5% methanol (saturated with ammonia) in dichloromethane affords 300 mg of the title product.

Physical characteristics are as follows:

FAB-HRMS: (m + H) = 527.3004.

Example 2 cyclo-(Pro-Phe-N-MeHis)-NHMe or L-histidinamide, L-prolyl-L-phenylalanyl-1-(carboxymethyl)-N,Nα-dimeth-yl-, cyclic (3→1)-peptide (Formula B-5: refer to chart B.)

To a stirred solution of 85 mg of H-Pro-Phe-N-MeHis-NHMe, which is the de-Boc'd title product of Preparation 4, and 0.04 ml of pyridine in 10 ml of dichloromethane is added dropwise a solution of 0.02 ml of bromoacetyl bromide in 10 ml of dichloromethane. After stirring overnight, the mixture is concentrated and then chromatographed on silica gel with 2%-5% methanol (saturated with ammonia) in dichloromethane to give 16 mg of the title product.

Physical characteristics are as follows:

FAB-HRMS: (m + H) = 508.2251.

15% renin inhibition at 10^{-7} M.

Preparation 5 Boc-Ala-N-MeHis(Ts)-NHMe (Formula C-1: refer to chart C.)

A solution of 380 mg of Boc-N-MeHis(Ts)-NHMe in 1 ml of dichloromethane and 1 ml of trifluoroacetic acid is allowed to stir for 45 minutes. The mixture is then added to a solution of 1.5 g of sodium bicarbonate in 15 ml of water. The resulting mixture is extracted with several portions of dichloromethane. The combined organic

phase is dried (magnesium sulfate) and then concentrated to give the free amine.

To a stirred solution of this residue and 250 mg of Boc-Ala-OH in 5 ml of dichloromethane is added 0.3 ml of diisopropylethylamine, followed by 350 mg of bis(2-oxo-3-oxazolidinyl)phosphinic chloride. The reaction mixture is stirred overnight, then flash chromatographed on silica gel with ethyl acetate to 3% methanol in ethyl acetate to afford 346 mg of the title product. The proposed structure is supported by ^{1}H-NMR.

Preparation 6 Boc-Pro-Ala-N-MeHis(Ts)-NHMe (Formula C-2: refer to Chart C.)

A solution of 346 mg of the title product of Preparation 5 in 1 ml of dichloromethane and 1 ml of trifluoroacetic acid is allowed to stir for 45 minutes. The mixture is then added to a solution of 1.5 g of sodium bicarbonate in 15 ml of water. The resulting mixture is extracted with several portions of dichloromethane. The combined organic phase is dried (magnesium sulfate) and then concentrated to give the free amine.

To a stirred solution of this residue and 183 mg of Boc-Pro-OH in 4 ml of dichloromethane is added 190 μl of diisopropylethylamine, followed by 135 μl of diethylphosphoryl cyanide. The reaction mixture is stirred overnight, then flash chromatographed on cilica with 5%-7% methanol in ethyl acetate to afford 372 mg of the title product. The proposed structure is supported by ^{1}H-NMR.

Preparation 7 Boc-Pro-Ala-N-MeHis-NHMe (Formula C-3: refer to Chart C.)

A solution of 372 mg of the title product of Preparation 6 and 376 mg of 1-hydroxybenzotriazole monohydrate in 2 ml of methanol is allowed to stand overnight and is then concentrated under reduced pressure. Chromatography of the residue on silica gel with 6% methanol (saturated with ammonia) in dichloromethane affords 269 mg of the title product.

Physical characteristics are as follows:

FAB-HRMS: (m + H) = 451.2664.

Example 3 cyclo-(Pro-Ala-N-MeHis)-NHMe or L-histidinamide, L-prolyl-L-alanyl-1-(carboxymethyl)-N,Nα-dimethyl-, cyclic (3→1)-peptide (Formula C-4: refer to Chart C.)

To a stirred solution of 130 mg of H-Pro-Ala-N-MeHis-NHMe, the de-Boc'd title product of Preparation 7, and 0.07 ml of pyridine in 20 ml of dichloromethane is added dropwise a solution of 0.035 ml of bromoacetyl bromide in 5 ml of dichloromethane. After stirring overnight, the mixture is concentrated and then chromatographed on silica

gel with 2%-5% methanol (saturated with ammonia) in dichloromethane to give 5 mg of the title product.

Physical characteristics are as follows:

FAB-HRMS: (m + H) = 390.2018.

HPLC (100% E) k' = 4.5.

IC_{50}: 5.5×10^{-7} M.

$K_i = 0.1\mu M$.

Preparation 8 Boc-Pro-Ala-N-MeHis-(CH_2CO_2tBu)-NHMe (Formula C-5: refer to Chart C.)

To a stirred solution of 252 mg of the title product of Preparation 7 in 3 ml of dichloromethane is added 0.2 ml of diisopropylethylamine, followed by 0.095 ml of tert-butylbromoacetate. After stirring at room temperature overnight, the mixture is concentrated and the residue chromatographed on silica gel with 2%-4% methanol (saturated with ammonia) in dichloromethane. The pooled material is then dried at 50°C in vacuo to give 300 mg of the title product.

Physical characteristics are as follows:

FAB-HRMS: (m + H) = 565.3346.

Preparation 9 H-Pro-Ala-N-MeHis(CH_2CO_2H)-NHMe·2TFA (Formula C-6: refer to Chart C.)

A solution of 156 mg of the title product of Preparation 8 in 0.3 ml of trifluoroacetic acid is allowed to stand at room temperature for one hour. This solution is slowly added to a vigorously stirred 20 ml solution of 2:1 hexane:ether. The precipitate is centrifuged and the supernatant removed. The residue is washed twice with 20 ml portions of 2:1 hexane:ether by centrifugation and supernatant removal. The resulting residue is then dried in vacuo to give 175 mg of a white solid.

Physical characteristics are as follows:

FAB-HRMS: (m + H) = 409.2181.

Example 4 cyclo-(Pro-Ala-N-MeHis)-NHMe or L-histidinamide, L-prolyl-L-alanyl-1-(carboxymethyl)-N,Nα-dimethyl-, cyclic (3→1)-peptide (Formula C-4: refer to Chart C.)

To a stirred suspension of 117.6 mg of the title product of Prepation 9 in 90 ml of dichloromethane is added 113 mg of dimethylaminopyridine, followed by 163.6 mg of the benzotriazol-1-yloxytris(dimethylamino)phosphonium hexafluorophosphate reagent.

After stirring at room temperature for 2.5 days, the solution is concentrated and the resulting residue is chromatographed on silica gel with 5%-8% methanol (saturated with ammonia) in dichloromethane to give 30 mg of the title product.

Physical characteristics are as given above in Example 3.

Preparation 10 Boc-His(Ts)-NHMe (Formula D-1: refer to Chart D.)

To a stirred solution of 1.0 g of Boc-His(Ts)-OH and 330 mg of methylamine hydrochloride in 10 ml of dichloromethane is added 1.53 ml of diisopropylethylamine, followed by 0.39 ml of diethylphosphoryl cyanide. The reaction mixture is stirred overnight, then flash chromatographed on silica with ethyl acetate to afford 672 mg of the title product. The proposed structure is supported by ^{1}H-NMR.

Preparation 11 Boc-Ala-His(Ts)-NHMe (Formula D-2: refer to Chart D.)

A solution of 390 mg of the title product of Preparation 10 in 1 ml of dichloromethane and 1 ml of trifluoroacetic acid is allowed to stir for 45 minutes. The mixture is then added to a solution of 1.5 g of sodium bicarbonate in 15 ml of water. The resulting mixture is extracted with several portions of dichloromethane. The combined organic phase is dried (magnesium sulfate) and then concentrated to give the free amine.

To a stirred solution of this residue and 210 mg of Boc-Ala-OH in 5 ml of dichloromethane is added 0.25 ml of diisopropylethylamine, followed by 0.18 ml of diethylphosphoryl cyanide. The reaction mixture is stirred overnight, then flash chromatographed on silica gel with ethyl acetate to 3% methanol in ethyl acetate to afford 352 mg of the title product. The proposed structure is supported by ^{1}H-NMR.

Preparation 12 Boc-Pro-Ala-His(Ts)-NHMe (Formula D-3: refer to Chart D.)

A solution of 352 mg of the title product of Preparation 11 in 1 ml of dichloromethane and 1 ml of trifluoroacetic acid is allowed to stir for 45 minutes. The mixture is then added to a solution of 1.5 g of sodium bicarbonate in 15 ml of water. The resulting mixture is extracted with several portions of dichloromethane. The combined organic phase is dried (magnesium sulfate) and then concentrated to give the free amine.

To a stirred solution of this residue and 190 mg of Boc-Pro-OH in 4 ml of dichloromethane is added 200 μl of diisopropylethylamine, followed by 140 μl of diethylphosphoryl cyanide. The reaction mixture is stirred overnight, then flash chromatographed on silica gel with 5%-7% methanol in ethyl acetate to afford 352 mg of the title product. The proposed structure is supported by ^{1}H-NMR.

Preparation 13 Boc-Pro-Ala-His-NHMe (Formula D-4: refer to Chart D.)

A solution of 352 mg of the title product of preparation 12 and 365 mg of 1-hydroxybenzotriazole monohydrate in 2 ml of methanol is allowed to stand overnight and is then concentrated under reduced pressure. Chromatography of the residue on silica gel with 6% methanol (saturated with ammonia) in dichloromethane affords 247 mg of the title product.

Physical characteristics are as follows:

FAB-HRMS: (m + H) = 437.2511.

Preparation 14 Boc-Pro-Ala-His(CH_2CO_2tBu)-NHMe (Formula D-5: refer to Chart D.)

To a stirred solution of 135 mg of the title product of Preparation 13 in 2 ml of dichloromethane and 0.5 ml of dimethylformamide is added 0.12 ml of diisopropylethylamine, followed by 0.055 ml of tert-butylbromoacetate. After stirring at room temperature overnight, the mixture is concentrated and the residue chromatographed on silica gel with 2%-4% methanol (saturated with ammonia) in dichloromethane. The pooled material is then dried at 50°C in vacuo to give 146 mg of the title product.

Physical characteristics are as follows:

FAB-HRMS: (m + H) = 551.3205.

Preparation 15 H-Pro-Ala-His(CH_2CO_2H)-NHMe·2TFA (Formula D-6: refer to Chart D.)

A solution of 146 mg of the title product of Preparation 14 in 0.3 ml of trifluoroacetic acid is allowed to stand at room temperature for one hour. This solution is slowly added to a vigorously stirred 20 ml of 2:1 hexane:ether. The precipitate is centrifuged and the supernatant removed. The residue is washed twice with 20 ml portions of 2:1 hexane:ether by centrifugation and supernatant removal. The resulting residue is then dried in vacuo to give 170 mg of a white solid.

Physical characteristics are as follows:

FAB-HRMS: (m + H) = 395.2042.

Example 5 cyclo-(Pro-Ala-His)-NHMe or L-histidinamide, L-prolyl-L-alanyl-1-(carbo ymethyl)-N-methyl-, cyclic (3→1)-peptide (Formula D-7: refer to Chart D.)

To a stirred suspension of 62 mg of the title product of Preparation 15 in 10 ml of dichloromethane is added 50 mg of dimethylaminopyridine, followed by 55 mg of the benzotriazol-1-yloxytris(dimethylamino)phosphoniumhexafluorophosphate. Afterstirring

at room temperature overnight, the solution is concentrated and the resulting residue is chromatographed on silica gel with 5%-8% methanol (saturated with ammonia) in dichloromethane to give 22 mg of the title product.

Physical characteristics are as follows:

FAB-HRMS: (m + H) = 377.1924.

HPLC (100% E) k' = 3.7.

11% renin inhibition at 10^{-7} M.

Preparation 16 Boc-Pro-Phe-His(Ts)-OBn (Formula E-2: refer to Chart E.)

To a stirred solution of 1.42 g of Boc-Pro-Phe-OH acid and 3.9 mmol of de-Boc'd compound of formula E-1 in 10 ml of dichloromethane is added 0.75 ml of diisopropylethylamine, followed by 0.66 ml of diethylphosphoryl cyanide. The solution is stirred overnight and then concentrated under reduced pressure. Flash chromatography of the residue on silica gel with 75% ethyl acetate in dichloromethane provides 2.97 g of the title product as a white foam.

Physical characteristics are as follows:

FAB-HRMS: (m + H) = 744.3061.

Preparation 17 Boc-Pro-Phe-His-OBn (Formula E-3: refer to Chart E.)

A solution of 1.12 g of the title product of Preparation 16 and 608 mg of 1-hydroxybenzotriazole monohydrate in a small amount of methanol is allowed to stand overnight and is then concentrated under reduced pressure. The residue is then chromatographed on a silica gel column which has been slurry-packed with 2.5% methanolic ammonia in dichloromethane. Elution with 3%-5% methanol (no ammonia) in dichloromethane provides 927 mg of the title product.

Physical characteristics are as follows:

^{1}H-NMR and FAB-HRMS: (m + H) = 590.2992.

Preparation 18 Boc-Pro-Phe-N^{im}(tert-butoxycarbonylmethyl)His-OBn (Formula E-4: refer to Chart E.)

To a stirred solution of 1.5 mmol of the title product of Preparation 17 in 6 ml of dichloromethane is added 0.29 ml of diisopropylethylamine followed by 0.24 ml of tert-butyl bromoacetate. The solution is stirred overnight and then chromatographed on silica gel. The silica gel column is slurry-packed with 2.5% methanolic ammonia in dichloromethane and eluted with 3%-4% methanol (no ammonia) in dichloromethane to afford 755 mg of the title product.

Physical characteristics are as follows:

FAB-HRMS: (m + H) = 704.3657.

Preparation 19 H-Pro-Phe-N^{im}(carboxymethyl)His-OBn·2 TFA

(Formula E-5: refer to Chart E.)

A solution of 755 mg of the title product of Preparation 18 in 2.5 ml of trifluoroacetic acid is allowed to stand for 75 minutes, then is diluted with an equal volume of dichloromethane. This solution is dripped slowly into 180 ml of rapidly-stirred 1:2 ether:hexane. The precipitated solid is spun down in a centrifuge and is washed twice by reslurrying in ether-hexane and recentrifuging. The solid is dried under vacuum to obtain 848 mg of the title product.

Example 6 cyclo(Pro-Phe-His)-OBn or L-histidine, L-prolyl-L-phenylalanyl-1-(carboxymethyl), cyclic (3→1)-peptide, benzyl ester (Formula E-6: refer to Chart E.)

To a stirred suspension of 1.07 mmol of the title product of Preparation 19 in 700 ml of dichloromethane is added 655 mg of DMAP, followed by 949 mg of BOP reagent. The salt gradually dissolves over the course of about one hour. After 5 days the solvent is removed under reduced pressure and the residue chromatographed on silica gel with 3%-7% methanol in dichloromethane to give 511 mg of white solid. Rechromatography on silica with 2-8% methanol in dichloromethane removes some of the impurities.

Physical characteristics are as follows:

^{1}H-NMR: δ 2.7, 2.8, 3.0, 3.2, 4.6, 6.59, 7.56.

FAB-HRMS: (m + H) = 530.2434.

15% renin inhibition at 10^{-7} M.

Preparation 20 cyclo(Pro-Phe-His)-OH (Formula E-7: refer to Chart E.)

A mixture of 106 mg of the title product of Example 6, 189 mg of ammonium formate, and 110 mg of 5% palladium on carbon in 2 ml of dimethylformamide is stirred vigorously overnight under argon. The mixture is filtered through Celite with methanol rinses of the filter cake. The filtrate is concentrated under reduced pressure to provide 106 mg of the title product, which is used directly in subsequent coupling reactions.

Physical characteristics are as follows:

FAB-HRMS: (m + H) = 440.1961.

Preparation 21 Boc-Pro-Phe-(NMe)His-CVG (Formula F-2: refer to Chart F.)

A solution of 116 mg of Boc-Pro-Phe-(NMe)His(Ts)-CVG of formula F-1 and 53

mg of 1-hydroxybenzotriazole monohydrate in a small amount of methanol is allowed to stand overnight and is then concentrated under reduced pressure. Chromatography of the residue on silica gel with 3%-4% methanol (saturated with ammonia) affords 81.2 mg of the title product.

Physical characteristics are as follows:

FAB-HRMS: (m + H) = 739.4763.

Preparation 22 Boc-Pro-Phe-(NMe)N^{im}-tert-butoxycarbonylmethyl)His-CVG (Formula F-3: refer to Chart F.)

To a stirred solution of 78.3 mg of the title product of Preparation 21 in 1 ml of dichloromethane is added 20 μl of diisopropylethylamine, followed by 17 μl of tert-butyl bromoacetate. After 2 days the mixture is chromatographed on silica gel (column slurry-packed with 3% methanolic ammonia in dichloromethane and eluted with 3%-4% methanol in dichloromethane) to afford 73.7 mg of the title product.

Physical characteristics are as follows:

FAB-HRMS: (m + H) = 853.5422.

Preparation 23 H-Pro-Phe-(NMe)(N^{im}-carboxymethyl)His-CVG·2TFA (Formula F-4: refer to Chart F.)

A solution of 71.4 mg of the title product of Preparation 22 in a small amount of trifluoroacetic acid is allowed to stand for one hour and is then dripped slowly into 15 ml of rapidly-stirred 2:1 hexane-ether. The solid is spun down in a centrifuge and washed twice with ether hexane, and finally dried under vacuum to give 65 mg of the title compound as its bis trifluoroacetate salt.

Example 7 cyclo-[Pro-Phe-(NMe)His]-CVG or L-histidinamide, L-prolyl-L-phenylalanyl-1-(carboxymethyl)-N-[[1-(cyclohexylmethyl)-2,3-dihydroxy-5-methyl]hexyl]-Nα-methyl-, cyclic (3→1)-peptide, [1S-[1R*,2S*,3R*]]- (Formula F-5: refer to Chart F.)

To a stirred suspension of 65 mg of the title product of Preparation 23 in 50 ml of dichloromethane is added 43 mg of DMAP, followed by 62 mg of BOP reagent. After 2 days the solution is concentrated under reduced pressure and the residue chromatographed on silica gel with 3%-5% methanol in dichloromethane to give the desired product heavily contaminated with DMAP. The mixture is rechromatographed on silica gel with 3-4 methanolic ammonia in dichloromethane to provide 20.2 mg of clean title product.

Physical characteristics are as follows:

FAB-HRMS: (m + H) = 679.4157.

HPLC (60/40 E/F, 225 nM) k' = 18.3.

IC_{50}: 1.2 x 10^{-6} M.

64% inhibition at 0.25 μM.

Example 8 cyclo(Pro-Phe-His)-$NH(CH_2)_2$-c-C_6H_{11} or L-histidinamide, L-prolyl-L-phenylalanyl-1-(carboxymethyl)-N-(2-cyclohexylethyl), cyclic (3→1)-peptide

To a solution of 30.4 mg of cyclo(Pro-Phe-His)-OH of Preparation 20 in 0.7 ml of dimethylformamide is added 15 μl of commercial cyclohexylethylamine, 13 μl of diisopropylethylamine, and 14 μl of diethylphosphoryl cyanide. After 18 hours, the dimethylformamide is pumped off and the residue chromatographed on silica gel with 3%-10% methanol in dichloromethane to provide 19.1 mg of the title product.

Physical characteristics are as follows:

FAB-HRMS: (m + H) = 549.3211.

HPLC (50/50 E/F; 225 nM) k' = 7.2.

16% renin inhibition at 10^{-7} M.

Example 9 cyclo(Pro-Phe-His)-cyclohexyl-alaninol or L-histidinamide, L-prolyl-L-phenylalanyl-1-(carboxymethyl)-N-[(1-cyclohexylmethyl-2-hydroxy)ethyl], cyclic (3→1)-peptide, [1S]-

To a suspension of 30.8 mg of cyclo(Pro-Phe-His)-OH of Preparation 20 and 16 mg of 2(S)-amino-3-cyclohexyl-1-propanol hydrochloride, which is prepared by hydrogenation of commercially available L-phenylalaninol, in 0.7 ml of dimethylformamide is added 28 μl of diisopropylethylamine, 2 mg of DMAP, and 37 mg of BOP reagent. A clear solution soon results, which is stirred overnight. Dimethylformamide is then pumped off and the residue chromatographed on silica gel with 3%-9% methanolic ammonia in dichloromethane to provide 25.0 mg of the title product.

Physical characteristics are as follows:

FAB-HRMS: (m + H) = 579.3285.

HPLC (60/40 E/F; 225 nM) k' = 6.9.

IC_{50}: 2.4 x 10^{-6} M.

Example 10 cyclo(Pro-Phe-His)-cyclohexylalanine, aldehyde or L-histidinamide, L-prolyl-L-phenylalanyl-1-(carboxymethyl)-N-[(1-cyclohexylmethyl-2-

oxo)ethyl], cyclic (3→1)-peptide, [1RS]-

To a cold (-78°C) stirred solution of 9 μl of oxalyl chloride in 0.2 ml of dry dichloromethane under argon is added 12 μl of dry dimethyl sulfoxide. The solution is stirred for 10 minutes at -78°C, then a solution of 30.5 mg of the title product of Example 9 in 0.4 ml of dichloromethane and 50 μl of dimethyl sulfoxide is added via a short cannula, with 2 small dichloromethane rinses. The solution is warmed to -30°C, stirred at that temperature for 20 minutes, then re-cooled to -78°C. After addition of 37 μl of diisopropylethylamine, the mixture is warmed to 0°C and given an aqueous workup. Thin layer chromatography at this point indicates substantial starting material remaining as well as the less polar product. Chromatography on silica gel with 5%-12% methanol in dichloromethane affords 15.2 mg of the title product.

Physical characteristics are as follows:

FAB-HRMS: (m + H) = 577.3143.

^{1}H-NMR: aldehyde doublet at δ 9.55.

HPLC (60/40 E/F, 225 nM) k' = 5.9.

8% renin inhibition at 10^{-8} M.

Example 11 cyclo-[Pro-Phe-His]-CVA-Mba or L-histidinamide, L-prolyl-L-phenylalanyl-1-(carboxymethyl)-N-[1-(cyclohexylmethyl)-2-hydroxy-5-methyl-4-[[[(2-methylbutyl)amino]carbonyl]hexyl], cyclic (3→1)-peptide, [1S-[1R*,2R*,4R*(2R*)]]-

To a stirred solution of 30.7 mg of acid cyclo(Pro-Phe-His)-OH of Preparation 20 and 31.6 mg of H-CVA-Mba hydrochloride prepared as described in PCT International Application PCT/US88/03436, Publication Number WO89/04833, published 1 June 1989, in 0.7 ml of dimethyl formamide is added 28 μl of diisopropylethylamine, followed by 13 μl of diethylphosphoryl cyanide. The solution is stirred overnight and then concentrated under reduced pressure. Chromatography of the residue on silica gel with 3%-8% methanolic ammonia in dichloromethane provides 9.2 mg of the title product.

Physical characteristics are as follows:

FAB-HRMS: (m + H) = 762.4912.

HPLC (50/50 E/F, 225 nM) k' = 12.6.

IC_{50}: 2.2 x 10^{-9} M.

Preparation 24 cyclo-[Pro-Phe-(NMe)His]-CVA(OTBS)-Ile-Amp

To a stirred solution of 36.6 mg of cyclo(Pro-Phe-His)-OH of Preparation 20 and 0.092 mmol of H-CVA(OTBS)-Ile-Amp prepared as described in PCT International Application PCT/US88/03436, Publication Number WO89/04833, published 1 June 1989, in 0.8 ml of dimethyl formamide is added 15 μl of diisopropylethylamine, followed by 2 ml of DMAP and 41 mg of BOP reagent. The solution is stirred overnight and then concentrated under reduced pressure. Chromatography of the residue on silica gel with 2%-5% methanolic ammonia in dichloromethane provides 55 mg of the title product.

Physical characteristics are as follows:

FAB-HRMS: (m + H) = 1010.623.

Example 12 cyclo-[Pro-Phe-His]-CVA-Ile-Amp or L-histidinamide, L-prolyl-L-phenylalanyl-1-(carboxymethyl)-N-[1-(cyclohexylmethyl)-2-hydroxy-5-methyl-4-[[[2-methyl-1-[[(2-pyridinylmethyl)amino]carbonyl]butyl]amino-]carbonyl]hexyl], cyclic (3→1)-peptide, [1S-[1R*,2R*,4R*(1R*, 2R*)]]-

A solution of 52.9 mg of the title product of Preparation 24 in 0.5 ml of 1:1 trifluoroacetic acid-dichloromethane is allowed to stand at room temperature. After 5 minutes, thin layer chromatography shows almost exclusively starting material, so the stopper is removed from the flask to allow evaporation of the dichloromethane component. Desilation appears complete after 4 hours, at which time the reaction mixture is diluted with dichloromethane and added to saturated aqueous bicarbonate. An insoluble solid is filtered off, and the organic layer dried (magnesium sulfate) and concentrated under reduced pressure. Chromatography of the residue on silica gel with 3%-9% methanolic ammonia in dichloromethane affords 36.9 mg of the title product.

Physical characteristics are as follows:

FAB-HRMS: (m + H) = 896.5384.

HPLC (10/90 E/F, 254 nM) k' = 19.9.

IC_{50}: 2.8×10^{-10} M.

91% inhibition at 0.25 μM.

Examples 13 - 16

Using procedures analogous to those described above and using starting materials and reactants readily known and available to one of ordinary skill in the art, the following additional compounds of the present invention, having the indicated physical

characteristics, are prepared:

(13) cyclo(Pro-Ala-His)-CVA-Ile-Amp;or L-histidinamide, L-prolyl-L-alanyl-1-(carboxymethyl)-N-[1-(cyclohexylmethyl)-2-hydroxy-5-methyl-4-[[[2-methyl-1-[[(2-pyridinylmethyl)amino]carbonyl]butyl]amino]carbonyl]hexyl], cyclic (3→1)-peptide, [1S-[1R*,2R*,4R*(1R*,2R*)]]-:

FAB-HRMS: (m + H) = 820.5071.

HPLC (50/50 E/F; 225 nM) k' = 6.8.

25% renin inhibition at 10^{-8} M.

(14) cyclo(Pro-Ala-His)-CVA-Mba; or L-histidinamide, L-prolyl-L-alanyl-1-(carboxymethyl)-N-[1-cyclohexylmethyl)-2-hydroxy-5-methyl-4-[[[(2-methylbutyl)amino]-carbonyl]hexyl], cyclic (3→1)-peptide, [1S-[1R*,2R*,4R*(2R*)]]-:

FAB-HRMS: (m + H) = 686.4608.

HPLC (50/50 E/F; 225 nM) k' = 5.0.

8% renin inhibition at 10^{-8} M.

(15) cyclo(Pro-Ala-His)-CVA-Ile-Amp→O;or L-histidinamide, L-prolyl-L-alanyl-1-(carboxymethyl)-N-[1-cyclohexylmethyl)-2-hydroxy-5-methyl-4-[[[(2-methyl-1-[[(2-pyridinylmethyl, N-oxide)amino]carbonyl]butyl]amino]carbonyl]hexyl], cyclic (3→1)-peptide, [1S-[1R*,2R*, 4R*(1R*,2R*)]]-:

FAB-HRMS: (m + H) = 836.5027.

HPLC (70/30 E/F; 225 nM) k' = 8.9.

18% renin inhibition at 10^{-8} M.

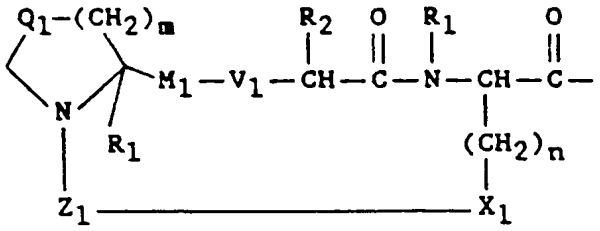
(16) cyclo(Pro-Ala-His)-CVG; or L-histidinamide, L-prolyl-L-alanyl-1-(carboxymethyl)-N-[[1-(cyclohexylmethyl)-2,3-dihydroxy-5-methyl]hexyl], cyclic (3→1)-peptide, [1S-[1R*,2S*,3R*]]-:

FAB-HRMS: (m + H) = 589.3723.

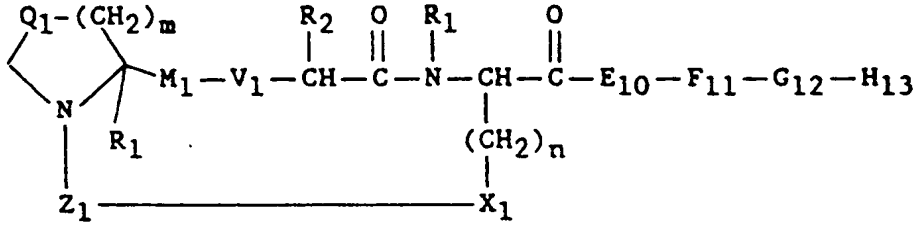
HPLC (70/30 E/F; 225 nM) k' = 9.6.

7% renin inhibition at 10^{-8} M.

FORMULA CHART



I



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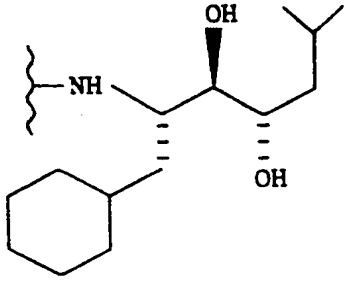

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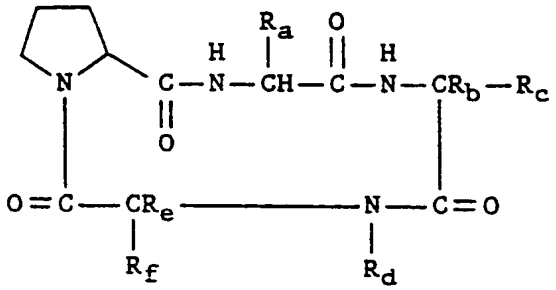

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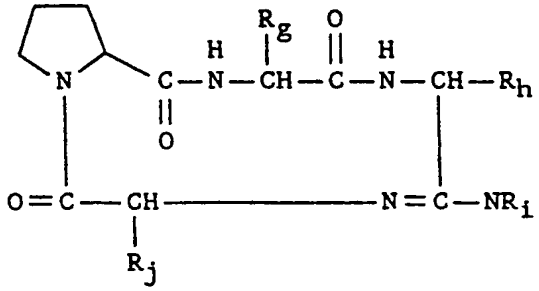

XL

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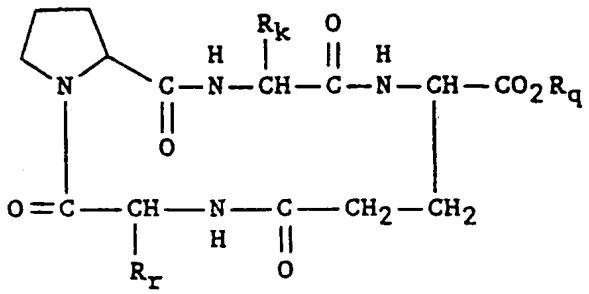
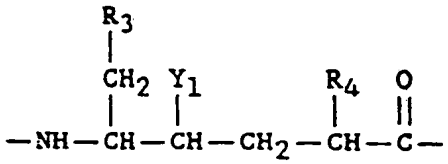
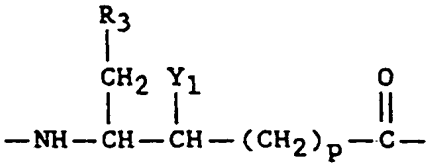


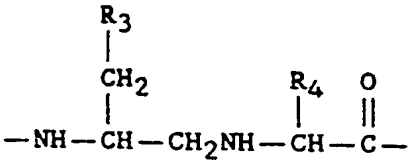
L



L_1



L_2



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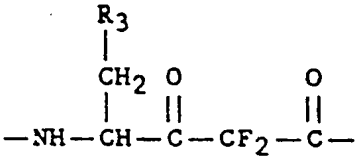
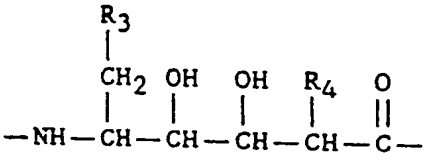


L_4



L_5

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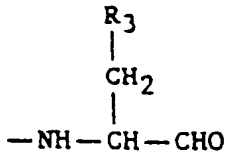
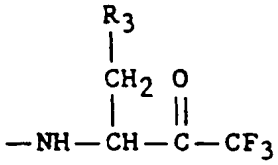
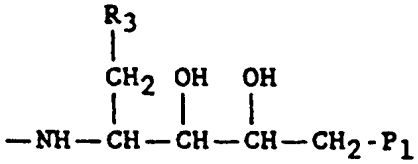


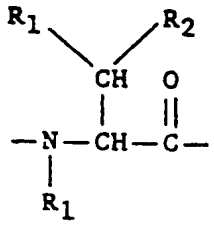
L6



L_7



L_8



L_9

CHART A

H-Pro-Phe-N-MeHis-LVA-Ile-Amp A-1

cyclo-(Pro-Phe-N-MeHis)-LVA-Ile-Amp A-2

CHART B

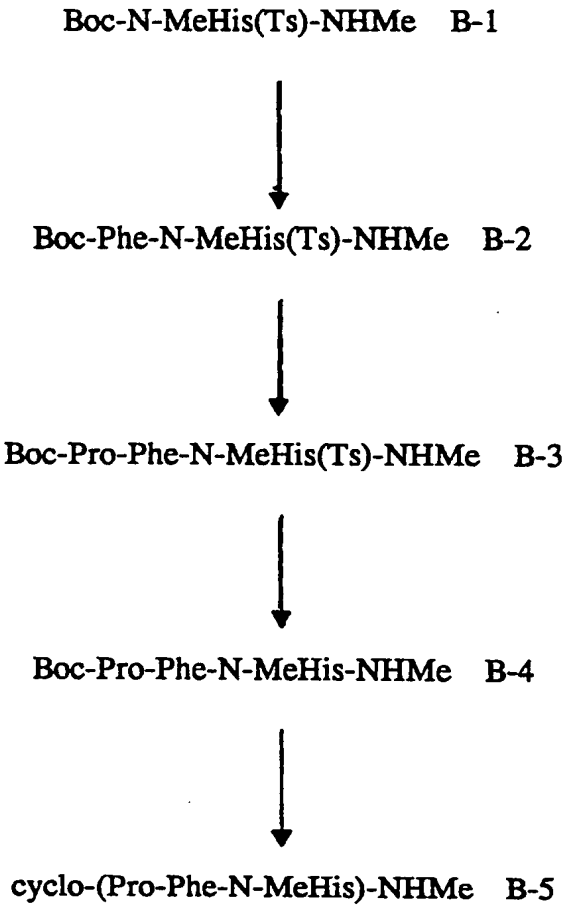

Boc-N-MeHis(Ts)-NHMe B-1
Boc-Phe-N-MeHis(Ts)-NHMe B-2
Boc-Pro-Phe-N-MeHis(Ts)-NHMe B-3
Boc-Pro-Phe-N-MeHis-NHMe B-4
cyclo-(Pro-Phe-N-MeHis)-NHMe B-5

CHART C

(Scheme 1)

Boc-Ala-N-MeHis(Ts)-NHMe C-1

Boc-Pro-Ala-N-MeHis(Ts)-NHMe C-2

Boc-Pro-Ala-N-MeHis-NHMe C-3

cyclo-(Pro-Ala-N-MeHis)-NHMe C-4

(Scheme 2)

Boc-Pro-Ala-N-MeHis-NHMe C-3

Boc-Pro-Ala-N-MeHis(CH_2CO_2tBu)-NHMe C-5

H-Pro-Ala-N-MeHis(CH_2CO_2H)-NHMe·2 TFA C-6

cyclo-(Pro-Ala-N-MeHis)-NHMe C-4

CHART D

Boc-His(Ts)-NHMe D-1

↓

Boc-Ala-His(Ts)-NHMe D-2

↓

Boc-Pro-Ala-His(Ts)-NHMe D-3

↓

Boc-Pro-Ala-His-NHMe D-4

↓

Boc-Pro-Ala-His(CH_2CO_2tBu)-NHMe D-5

↓

H-Pro-Ala-His(CH_2CO_2H)-NHMe·2 TFA D-6

↓

cyclo-(Pro-Ala-His)-NHMe D-7

CHART E

Boc-His(Ts)-OBn E-1

↓

Boc-Pro-Phe-His(Ts)-OBn E-2

↓

Boc-Pro-Phe-His-OBn E-3

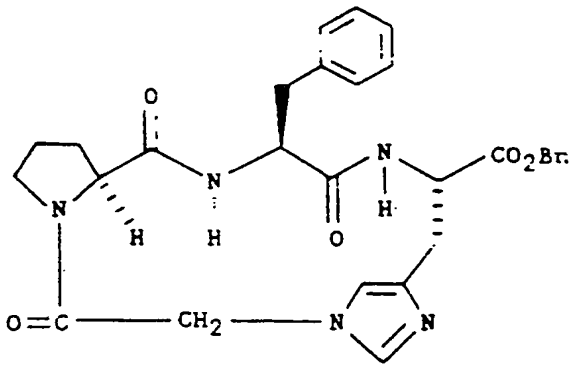
↓

Boc-Pro-Phe-His(CH_2CO_2tBu)-OBn E-4

↓

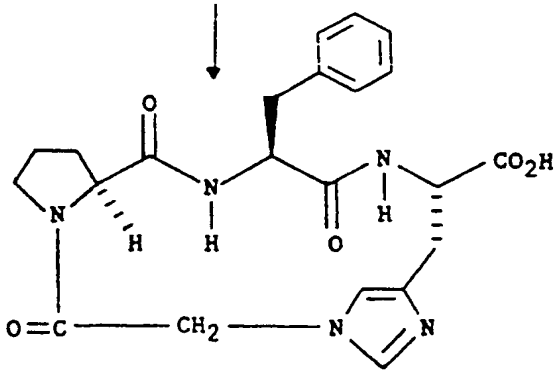
H-Pro-Phe-His(CH_2CO_2H)-OBn·2 TFA E-5

↓



E-6

↓

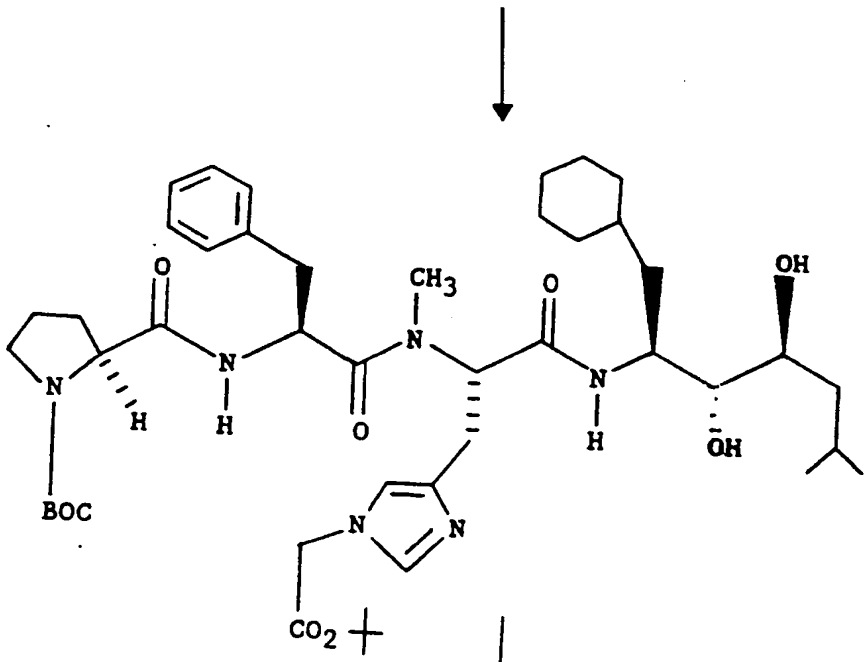


E-7

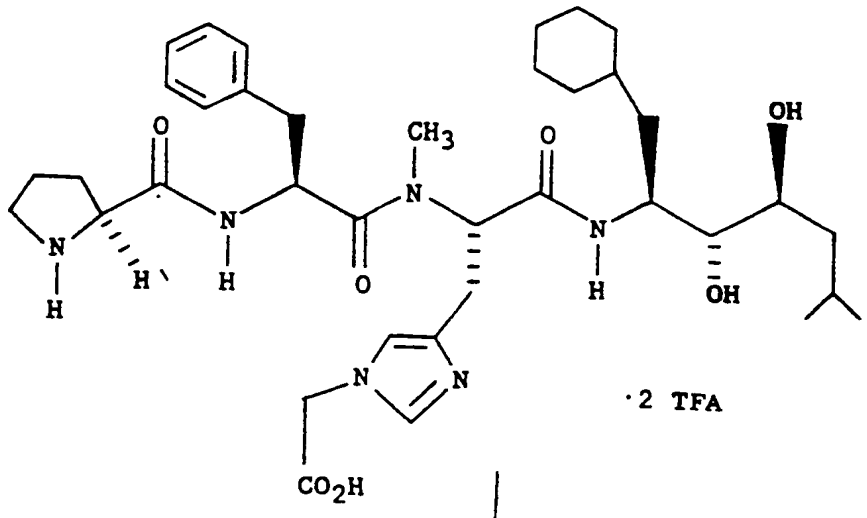
CHART F

Boc-Pro-Phe-(NMe)His(Ts)-CVG F-1

Boc-Pro-Phe-(NMe)His-CVG F-2



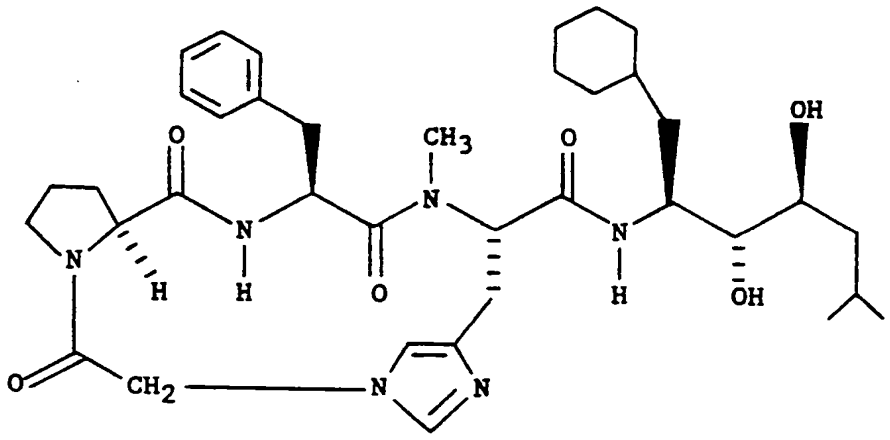
F-3



F-4

·2 TFA

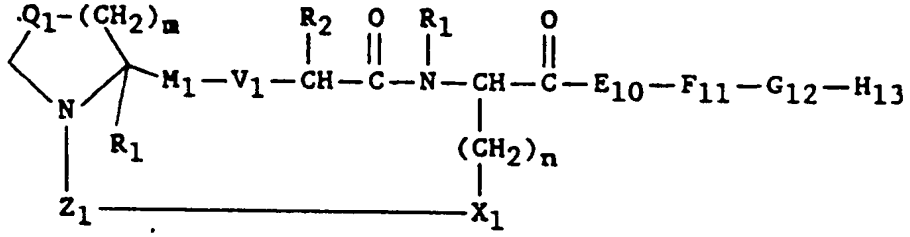
CHART F (continued)



F-5

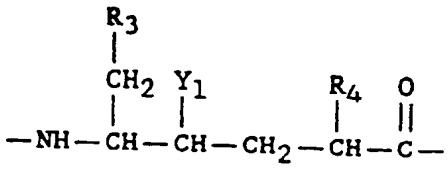
CLAIMS

1. A compound of formula II

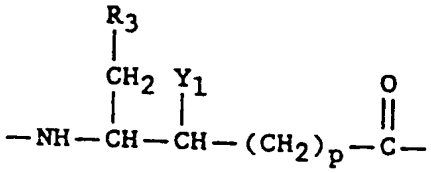


II

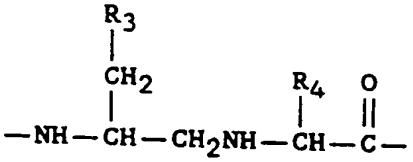
wherein E_{10}-F_{11} is absent or a divalent moiety of the formula L_1, L_2, L_3, L_4 or L_5, or a monovalent moiety of the formula L_6, L_7 or L_8;



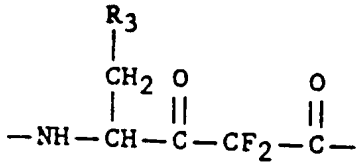
L_1



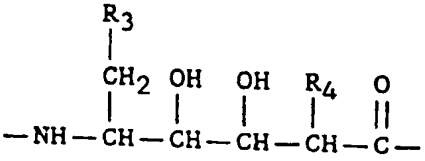
L_2



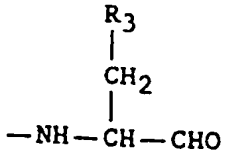
L_3



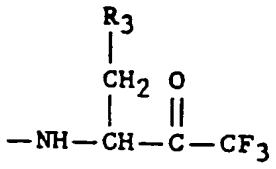
L_4



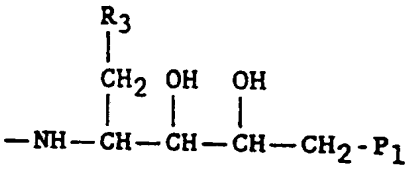
L_5



L_6

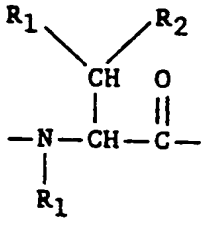


L_7



L_8

wherein G_{12} is absent or a divalent moiety of the formula L_9;



L_9

wherein H_{13} is

a) $-O-R_5$, or

b) $-N(R_1)(R_5)$;

wherein Q_1 is

a) $-CH_2-$,

b) -CH(OH),

c) -O-, or

d) -S-;

wherein M_1 is

a) -C(O)-, or

b) $-CH_2-$;

wherein V_1 is

a) -O-, or

b) $-N(R_1)-$;

wherein X_1 is

a) -Het,

b) $-N(R_1)-$, or

c) -S-;

wherein Y_1 is

a) -OH, or

b) $-NH_2$;

wherein Z_1 is

a) $-C(O)-(CH_2)_p-$, or

b) $-(CH_2)_p-C(O)-$;

wherein P_1 is

a) $-N_3$,

b) -CN,

c) C_1-C_6alkyl,

d) C_1-C_6cycloalkyl,

e) aryl, or

f) het;

wherein m is one or two;

wherein n is one to five, inclusive;

wherein p is zero to five, inclusive;

wherein aryl is phenyl or naphthyl, optionally substituted by 0 to 3 of the following:

(a) C_1-C_5alkyl,

(b) hydroxy,

(c) hydroxy(C_1-C_5alkyl),

(d) C_1-C_5alkoxy,

(e) amino,

(f) amino(C_1-C_5alkyl),

(g) halogen,

(h) -CHO,

(i) $-CO_2H$,

(j) $-CO_2-(C_1-C_5alkyl)$,
(k) $-CONH_2$,
(l) $-CONH(C_1-C_5alkyl)$,
(m) nitro,
(n) mercapto,
(o) $mercapto(C_1-C_5alkyl)$,
(p) $-SO_2H$,
(q) $-SO_2NH_2$, or
(r) -CN;

wherein -Het is a 5- or 6-membered saturated or unsaturated ring containing from 1 to 3 heteroatoms (nitrogen, oxygen, or sulfur), and including any bicyclic group in which any of the above heterocyclic rings is fused to a benzene ring or another heterocycle, and if chemically feasible, the nitrogen and sulfur atoms may be in the oxidized forms; and optionally substituted by 0 to 3 of the following:

(a) C_1-C_5alkyl,
(b) hydroxy,
(c) $hydroxy(C_1-C_5alkyl)$,
(d) $C_1-C_5alkoxy$,
(e) amino,
(f) $amino(C_1-C_5alkyl)$,
(g) halogen,
(h) -CHO,
(i) $-CO_2H$,
(j) $-CO_2-(C_1-C_5alkyl)$,
(k) $-CONH_2$,
(l) $-CONH(C_1-C_5alkyl)$,
(m) nitro,
(n) mercapto,
(o) $mercapto(C_1-C_5alkyl)$,
(p) $-SO_2H$,
(q) $-SO_2NH_2$, or
(r) -CN;

wherein R_1 is

(a) hydrogen, or

(b) C_1-C_5alkyl;

wherein R_2 is

(a) hydrogen,

(b) C_1-C_8alkyl,

(c) -$(CH_2)_p$-aryl,

(d) -$(CH_2)_p$-het,

(e) -$(CH_2)_p$-CO_2H,

(f) -$(CH_2)_p$-NH_2,

(g) -$(CH_2)_p$-$CH(NH_2)(CO_2H)$,

(h) C_3-C_7cycloalkyl, or

(i) 1- or 2-adamantyl;

wherein R_3 is

(a) hydrogen,

(b) C_1-C_5alkyl,

(c) aryl,

(d) C_3-C_7cycloalkyl,

(e) het,

(f) C_1-C_3alkoxy, or

(g) C_1-C_3alkylthio;

wherein R_4 is

(a) hydrogen,

(b) C_1-C_8alkyl,

(c) C_3-C_7cycloalkyl, or

(d) -$CH(R_1)(R_6)$;

wherein R_5 is

(a) hydrogen,

(b) C_1-C_{10}alkyl,

(c) -(CH_2)p-alkyl,

(d) -$(CH_2)_p$-het,

(e) -$(CH_2)_p$-cycloalkyl,

(f) -$(CH_2)_p$-$CH(NH_2)(CO_2H)$, or

(g) -$(CH_2)_n$-R_7;

wherein R_6 is

(a) hydrogen,

(b) hydroxy,

(c) C_1-C_5alkyl,

(d) C_3-C_7cycloalkyl,

(e) aryl,

(f) -Het,

(g) C_1-C_3alkoxy, or

(h) C_1-C_3alkylthio;

wherein R_7 is

(a) hydroxy,

(b) amino,

(c) $-CO_2H$,

(d) $-SO_2H$,

(e) $-SO_2NH_2$,

(f) guadinyl, or

(g) a polyhydroxylated-substituted-alkyl moiety;

or a carboxy-, amino- or other reactive group protected form thereof;

or a pharmaceutically acceptable acid or base addition salt thereof; provided that:

1) when E_{10}-F_{11} is the moiety of formula L_6, L_7 or L_8, then G_{12} and H_{13} are absent; and

2) when X_1 is -Het, it is bonded to $-(CH_2)_n$ of formula II as chemically feasible and is bonded to Z_1 of formula II by a heteroatom of -Het as chemically feasible.

2. The compound of claim 1

wherein the moiety of formula I is

a) cyclo-(Pro-Phe-N-MeHis)-,

b) cyclo-(Pro-Ala-N-MeHis)-,

c) cyclo-(Pro-Ala-His)-, or

d) cyclo-(Pro-Phe-His)-;

wherein E_{10}-F_{11} is

a) absent,

b) LVA, or derivatives thereof,

c) CVA, or derivatives thereof,

d) CVG, or derivatives thereof, or

e) the moiety of formula L_6;

wherein G_{12} is

a) absent, or

b) Ile, or derivatives thereof;

wherein H_{13} is

a) absent,

b) Amp,

c) Amp→O,

d) -NH(CH_3),

e) -O-benzyl,

f) -NH((CH_2)$_2$-c-C_6H_{11}),

g) -Mba, or

h) cyclohexylalaninol.

3. The compound of claim 2 selected from the group consisting of:

Cyclo-(Pro-Phe-N-MeHis)-LVA-Ile-Amp or L-Histidinamide, L-prolyl-L-phenylalanyl-1-(carboxymethyl)-N-[2-hydroxy-5-methyl-1-(2-methylpropyl)-4-[[[2-methyl-1-[1(2-pyridinylmethyl)amino]carbonyl]butyl]amino]carbonyl]hexyl]-Nα-methyl-,cyclic (3→1)-peptide, [1S-[1R*,2R*,4R*(1R*,2R*)]]-;

Cyclo-(Pro-Phe-N-MeHis)-NHMe or L-Histidinamide, L-prolyl-L-phenylalanyl-1-(carboxymethyl)-N,Nα-dimethyl-, cyclic (3→1)- peptide; Cyclo-(Pro-Ala-N-MeHis)-NHMeorL-Histidinamide,L-prolyl-L-alanyl-1-(carboxymethyl)-N,Nα-dimethyl-, cyclic (3→1)-peptide

Cyclo-(Pro-Ala-His)-NHMe or L-Histidinamide, L-prolyl-L-alanyl-1-(carboxymethyl)-N-methyl-, cyclic (3→1)-peptide;

Cyclo-(Pro-Phe-(NMe)His)-CVG or L-Histidinamide, L-prolyl-L-phenylalanyl-1-(carboxymethyl)-N-[[1-(cyclohexylmethyl)-2,3-dihydroxy-5-methyl]hexyl]-Nα-methyl-, cyclic (3→1)-peptide, [1S-[1R*,2S*,3R*]]-;

Cyclo-(Pro-Phe-His)-OBn or L-Histidine, L-prolyl-L-phenylalanyl-1-(carboxymethyl), cyclic (3→1)-peptide, benzyl ester;

Cyclo-(Pro-Phe-His)-NH(CH_2)$_2$-c-C_6H_{11} or L-Histidinamide, L-prolyl-L-

phenylalanyl-1-(carboxymethyl)-N-(2-cyclohexylethyl), cyclic (3→1)-peptide;

Cyclo-(Pro-Phe-His)-CVA-Ile-Amp or L-Histidinamide, L-prolyl-L-phenylalanyl-1-(carboxymethyl)-N-[1-(cyclohexylmethyl)-2-hydroxy-5-methyl-4-[[[2-methyl-1-[[(2-pyridinylmethyl)amino]carbonyl]butyl]amino]carbonyl]hexyl],cyclic (3→1)-peptide, [1S-[1R*,2R*,4R*(1R*, 2R*)]]-;

Cyclo-(Pro-Phe-His)-CVA-Mba or L-Histidinamide, L-prolyl-L-phenylalanyl-1-(carboxymethyl)-N-[1-(cyclohexylmethyl)-2-hydroxy-5-methyl-4-[[[(2-methylbutyl-)amino]carbonyl]hexyl], cyclic (3→1)-peptide, [1S-[1R*,2R*,4R*(2R*)]]-;

Cyclo-(Pro-Phe-His)-cyclohexyl-alaninol or L-Histidinamide, L-prolyl-L-phenylalanyl-1-(carboxymethyl)-N-[(1-cyclohexylmethyl-2-hydroxy)ethyl], cyclic (3→1)-peptide, [1S]-;

Cyclo-(Pro-Phe-His)-cyclohexylalanine aldehyde or L-Histidinamide, L-prolyl-L-phenylalanyl-1-(carboxymethyl)-N-[(1-cyclohexylmethyl-2-oxo)ethyl], cyclic (3→1)-peptide, [1RS]-;

Cyclo-(Pro-Ala-His)-CVA-Ile-Amp or L-Histidinamide, L-prolyl-L-alanyl-1-(carboxymethyl)-N-[1-(cyclohexylmethyl)-2-hydroxy-5-methyl-4-[[[2-methyl-1-[[(2-pyridinylmethyl)amino]carbonyl]butyl]amino]carbonyl]hexyl],cyclic (3→1)-peptide, [1S-[1R*,2R*,4R*(1R*,2R*)]]-;

Cyclo-(Pro-Ala-His)-CVA-Mba or L-Histidinamide, L-prolyl-L-alanyl-1-(carboxymethyl)-N-[1-cyclohexylmethyl)-2-hydroxy-5-methyl-4-[[[(2-methylbutyl)amino]-carbonyl]hexyl], cyclic (3→1)-peptide, [1S-[1R*,2R*,4R*(2R*)]]-;

Cyclo-(Pro-Ala-His)-CVA-Ile-Amp→O or L-Histidinamide, L-prolyl-L-alanyl-1-(carboxymethyl)-N-[1-cyclohexylmethyl)-2-hydroxy-5-methyl-4-[[[(2-methyl-1-[[(2-pyridinylmethyl, N-oxide)amino]carbonyl]butyl]amino]carbonyl]hexyl], cyclic (3→1)-peptide, [1S-[1R*,2R*, 4R*(1R*,2R*)]]-; and

Cyclo-(Pro-Ala-His)-CVG or L-Histidinamide, L-prolyl-L-alanyl-1-(carboxy-methyl)-N-[[1-(cyclohexylmethyl)-2,3-dihydroxy-5-methyl]hexyl],cyclic (3→1)-peptide, [1S-[1R*,2S*,3R*]]-.

INTERNATIONAL SEARCH REPORT

International Application No PCT/US 90/05744

I. CLASSIFICATION OF SUBJECT MATTER (if several classification symbols apply, indicate all) [6]

According to International Patent Classification (IPC) or to both National Classification and IPC

IPC[5]: C 07 K 5/02, C 07 K 7/54, A 61 K 37/64

II. FIELDS SEARCHED

Minimum Documentation Searched [7]

Classification System	Classification Symbols
IPC[5]	C 07 K, A 61 K

Documentation Searched other than Minimum Documentation to the Extent that such Documents are included in the Fields Searched [8]

III. DOCUMENTS CONSIDERED TO BE RELEVANT [9]

Category [10]	Citation of Document, [11] with indication, where appropriate, of the relevant passages [12]	Relevant to Claim No. [13]
A	J. Med. Chem., vol. 31, 1988, American Chemical Society, H.L. Sham et al.: "Renin inhibitors. Design and synthesis of a new class of conformationally restricted analogues of angiotensinogen", pages 284-295 see pages 284-285; table II; page 291, discussion - page 292, column 1 --	1-3
P,X	Abstracts of Papers, 200th ACS National Meeting, Washington, 26-31 August 1990, part 1, MEDI 105, S. Thaisrivongs et al.: "Conformationally constrained renin inhibitory peptides: cyclic (3-1)-1-(carboxymethyl)-L-prolyl-L-phenylalanyl-L-histidinamide as conformational restriction at the P2-P4 tripeptide portion of the angiotensinogen template", see the whole abstract ----------	

* Special categories of cited documents: [10]

"A" document defining the general state of the art which is not considered to be of particular relevance

"E" earlier document but published on or after the international filing date

"L" document which may throw doubts on priority claim(s) or which is cited to establish the publication date of another citation or other special reason (as specified)

"O" document referring to an oral disclosure, use, exhibition or other means

"P" document published prior to the international filing date but later than the priority date claimed

"T" later document published after the international filing date or priority date and not in conflict with the application but cited to understand the principle or theory underlying the invention

"X" document of particular relevance; the claimed invention cannot be considered novel or cannot be considered to involve an inventive step

"Y" document of particular relevance; the claimed invention cannot be considered to involve an inventive step when the document is combined with one or more other such documents, such combination being obvious to a person skilled in the art.

"&" document member of the same patent family

IV. CERTIFICATION

Date of the Actual Completion of the International Search	Date of Mailing of this International Search Report
25th February 1991	18. 03. 91
International Searching Authority	**Signature of Authorized Officer**
EUROPEAN PATENT OFFICE	N. Toribio Nuria TORIBIO